

Supplementary Information for

Phylogenomic analysis of velvet worms (*Onychophora*) uncovers an evolutionary radiation in the Neotropics

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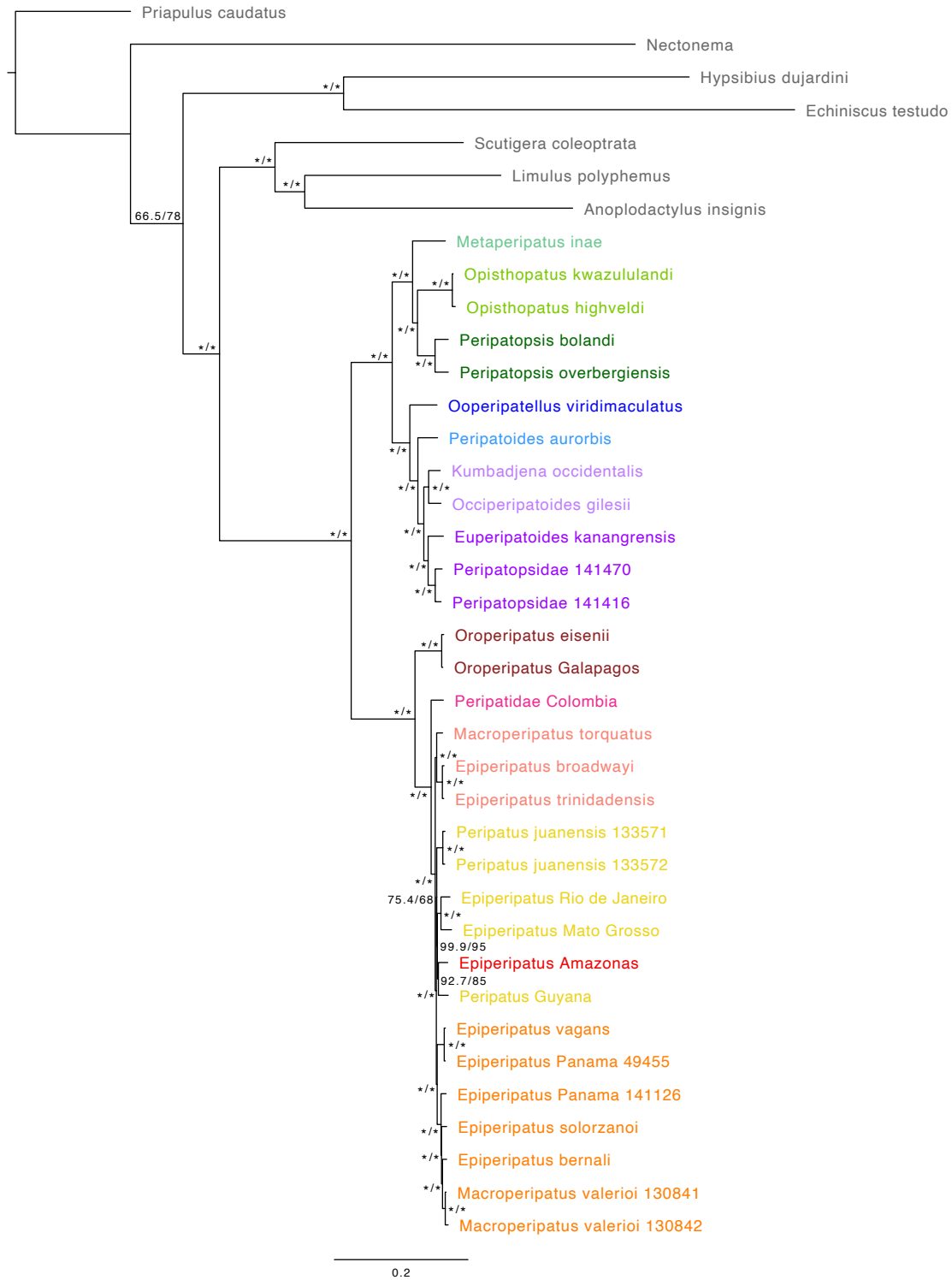


Figure S1. Phylogeny inferred from IQ-TREE analysis of M1. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

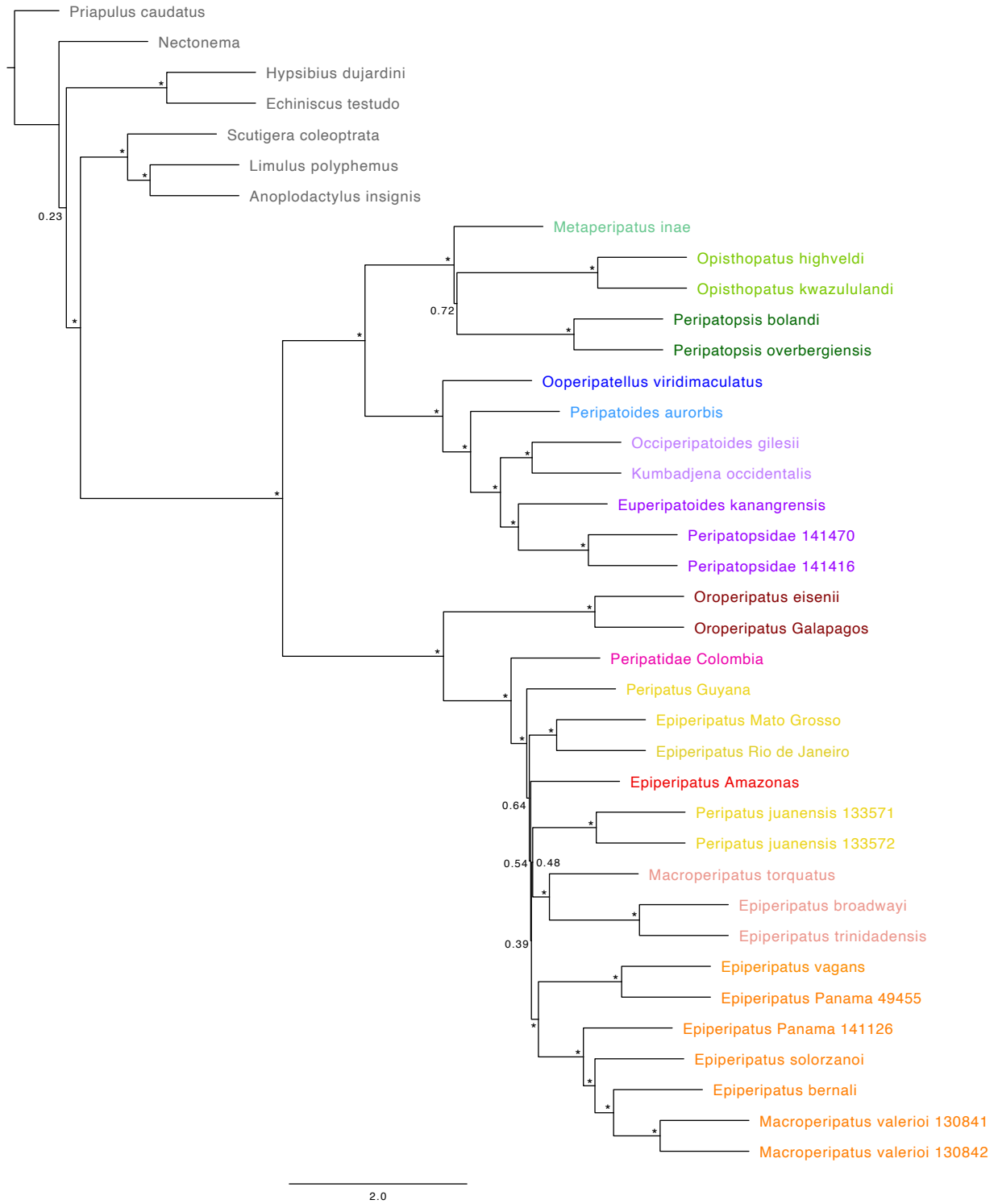


Figure S2. Phylogeny inferred from ASTRAL analysis of M1. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

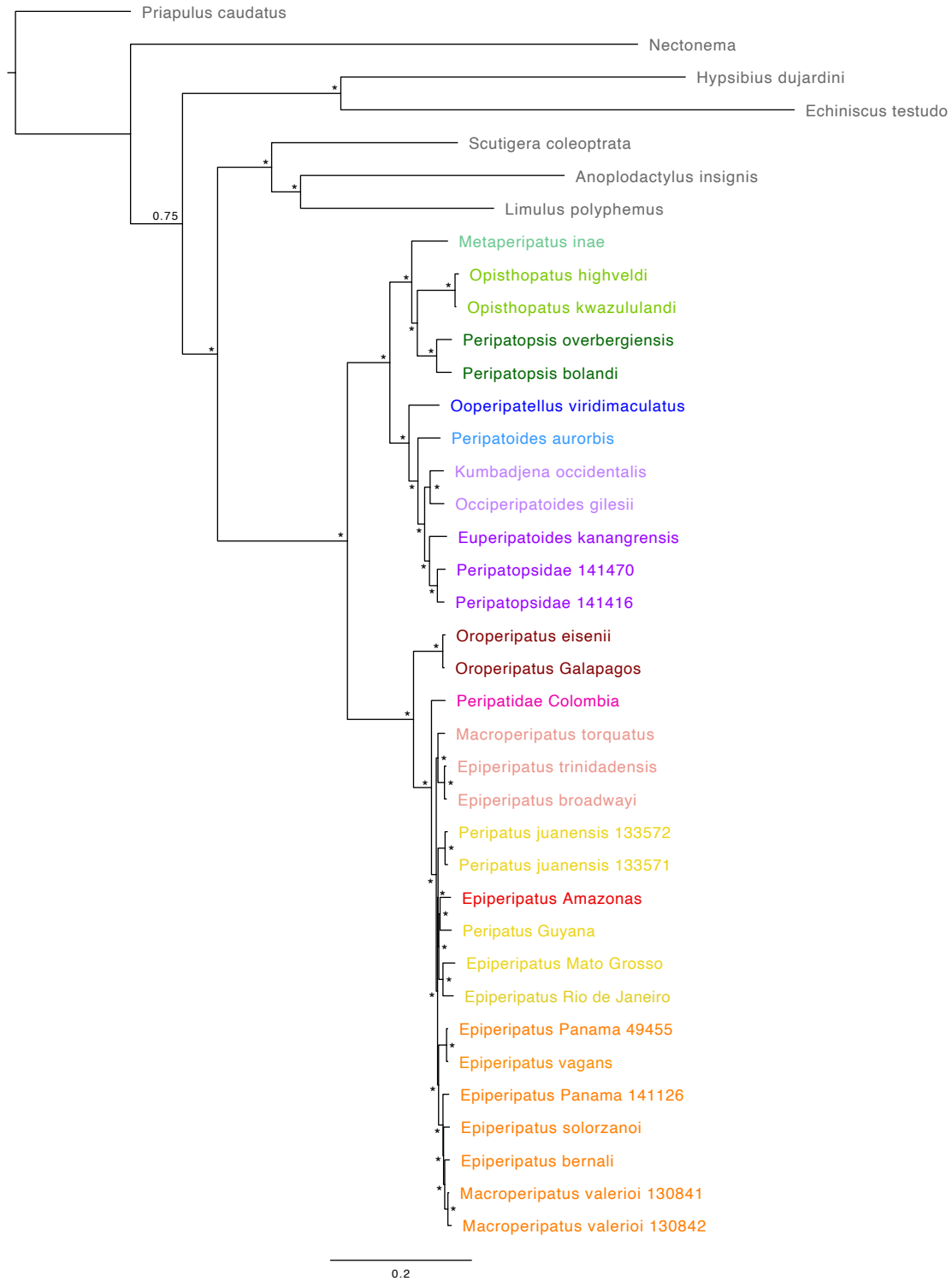


Figure S3. Phylogeny inferred from ExaBayes analysis of M1. Asterisks (*) mark nodes with full support; remaining nodes show the posterior probability.

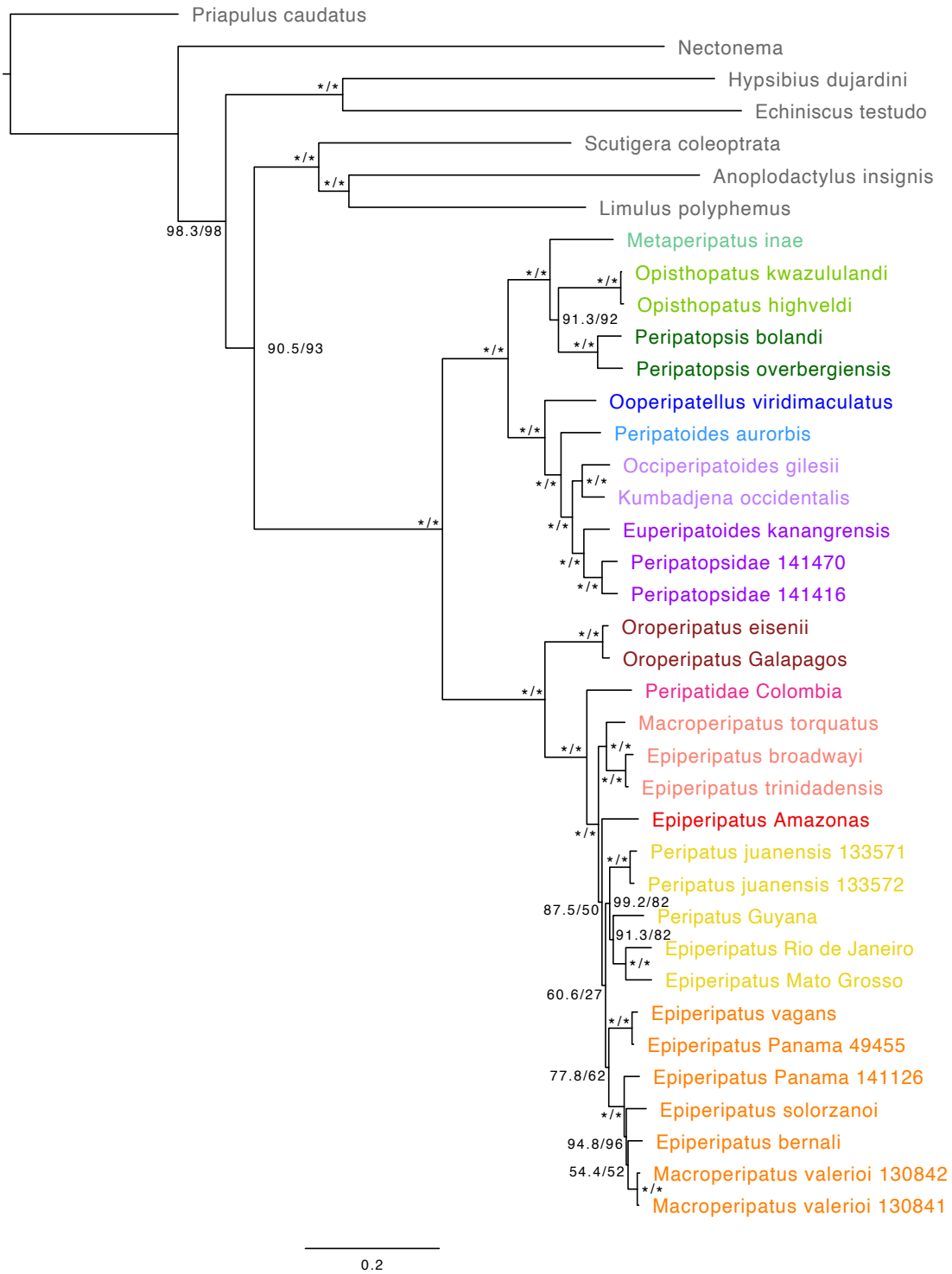


Figure S4. Phylogeny inferred from IQ-TREE analysis of M2. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

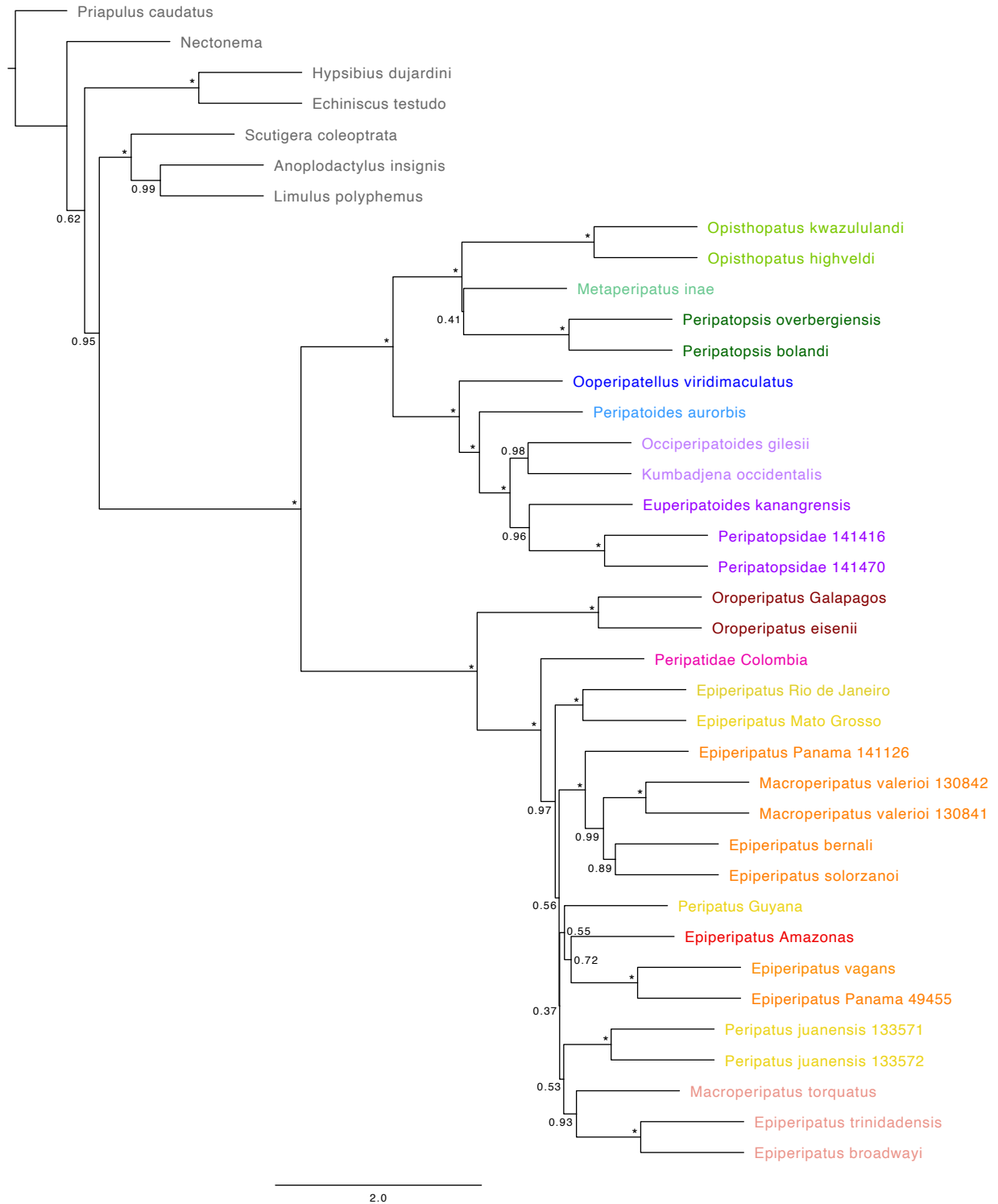


Figure S5. Phylogeny inferred from ASTRAL analysis of M2. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

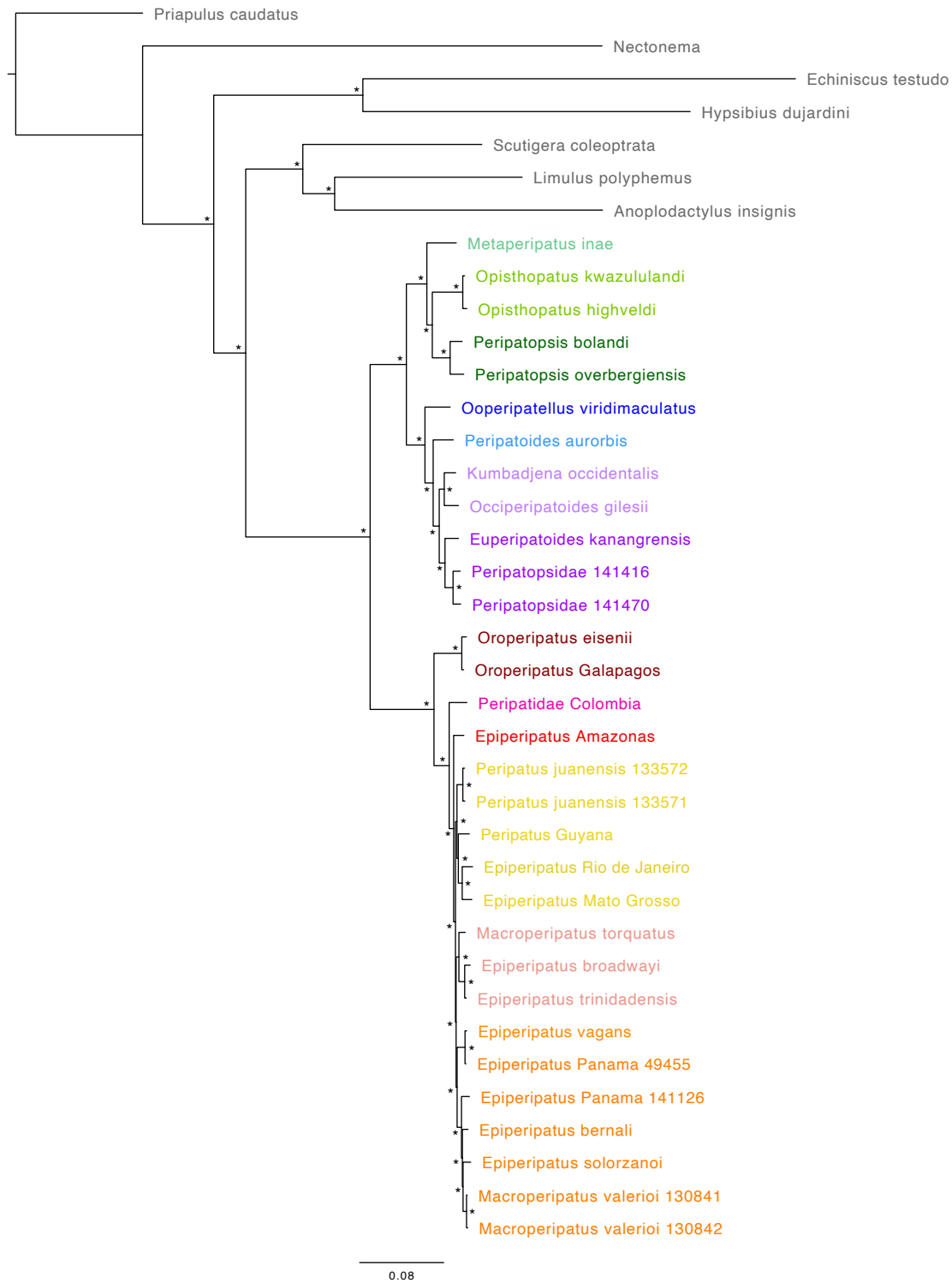


Figure S6. Phylogeny inferred from ExaBayes analysis of M2. Asterisks (*) mark nodes with full support.

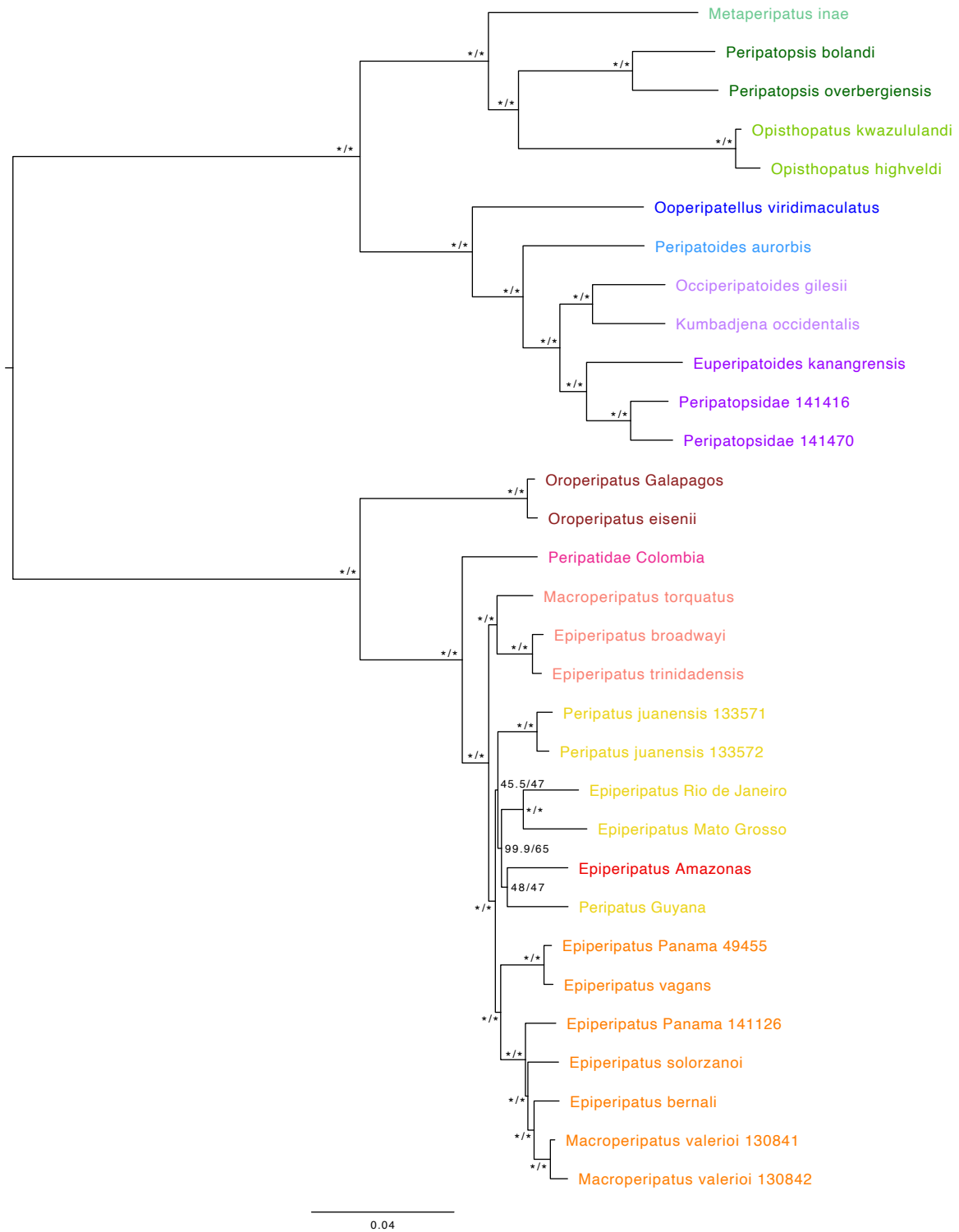


Figure S7. Phylogeny inferred from IQ-TREE analysis of M3. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

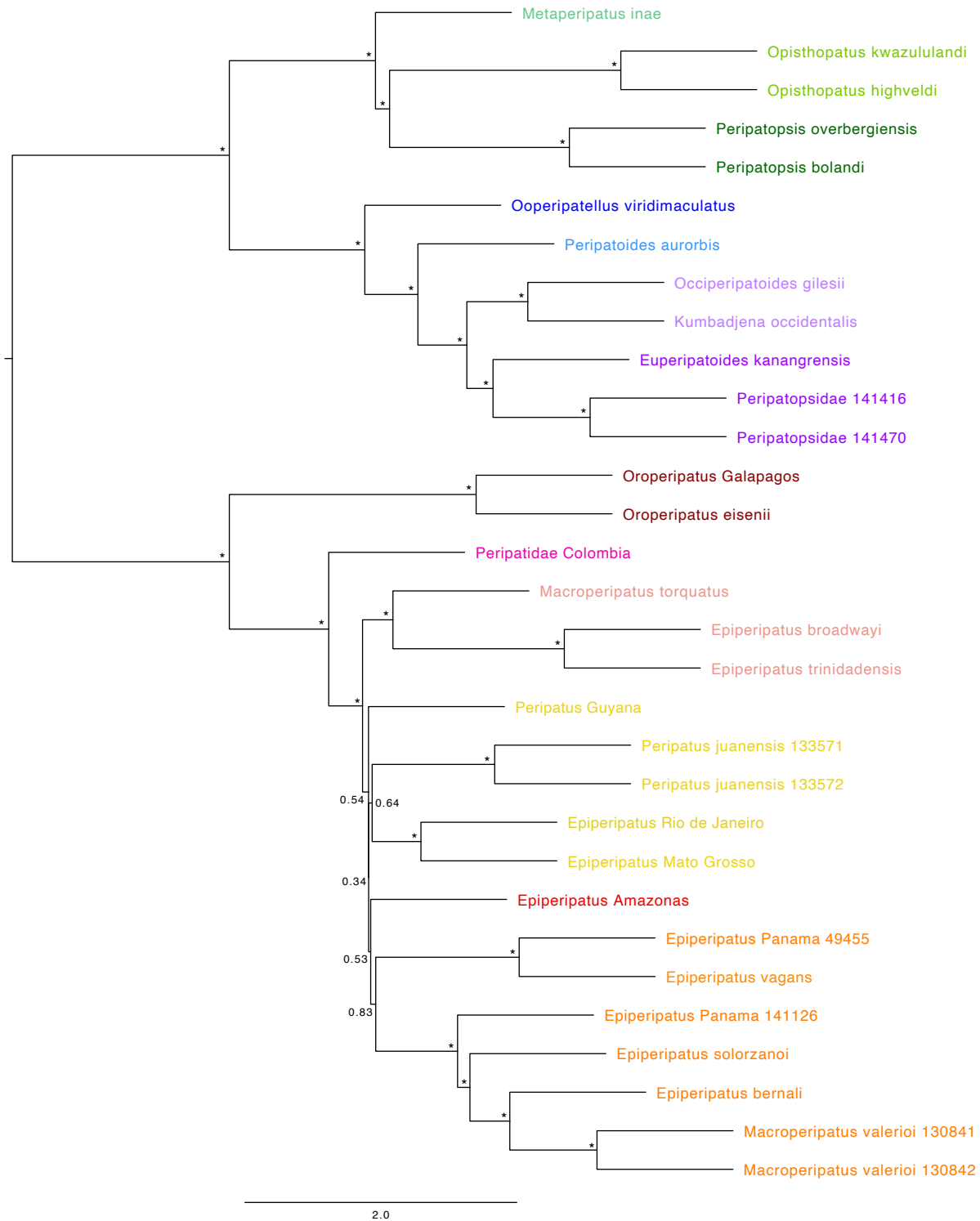


Figure S8. Phylogeny inferred from ASTRAL analysis of M3. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

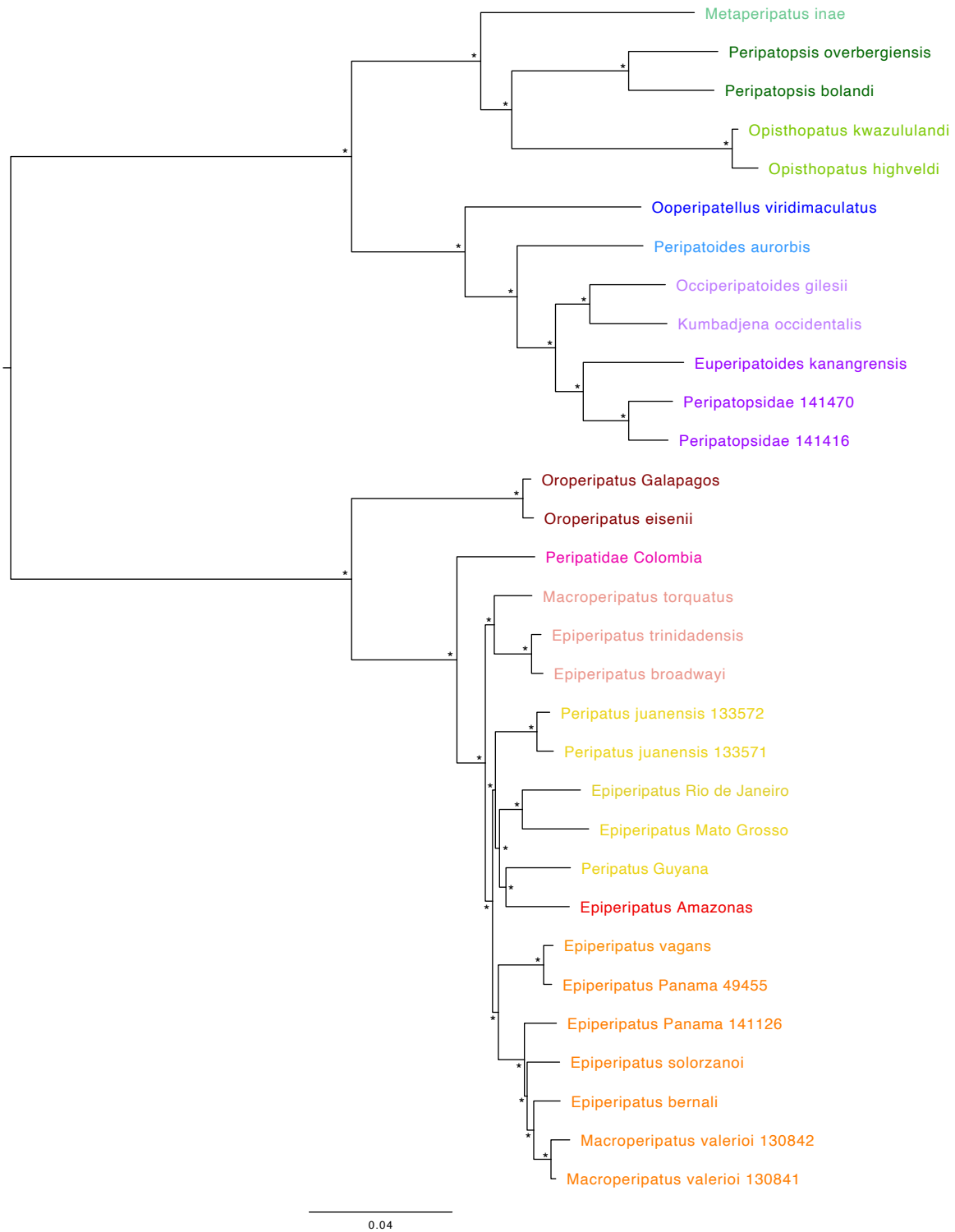


Figure S9. Phylogeny inferred from ExaBayes analysis of M3. Asterisks (*) mark nodes with full support.

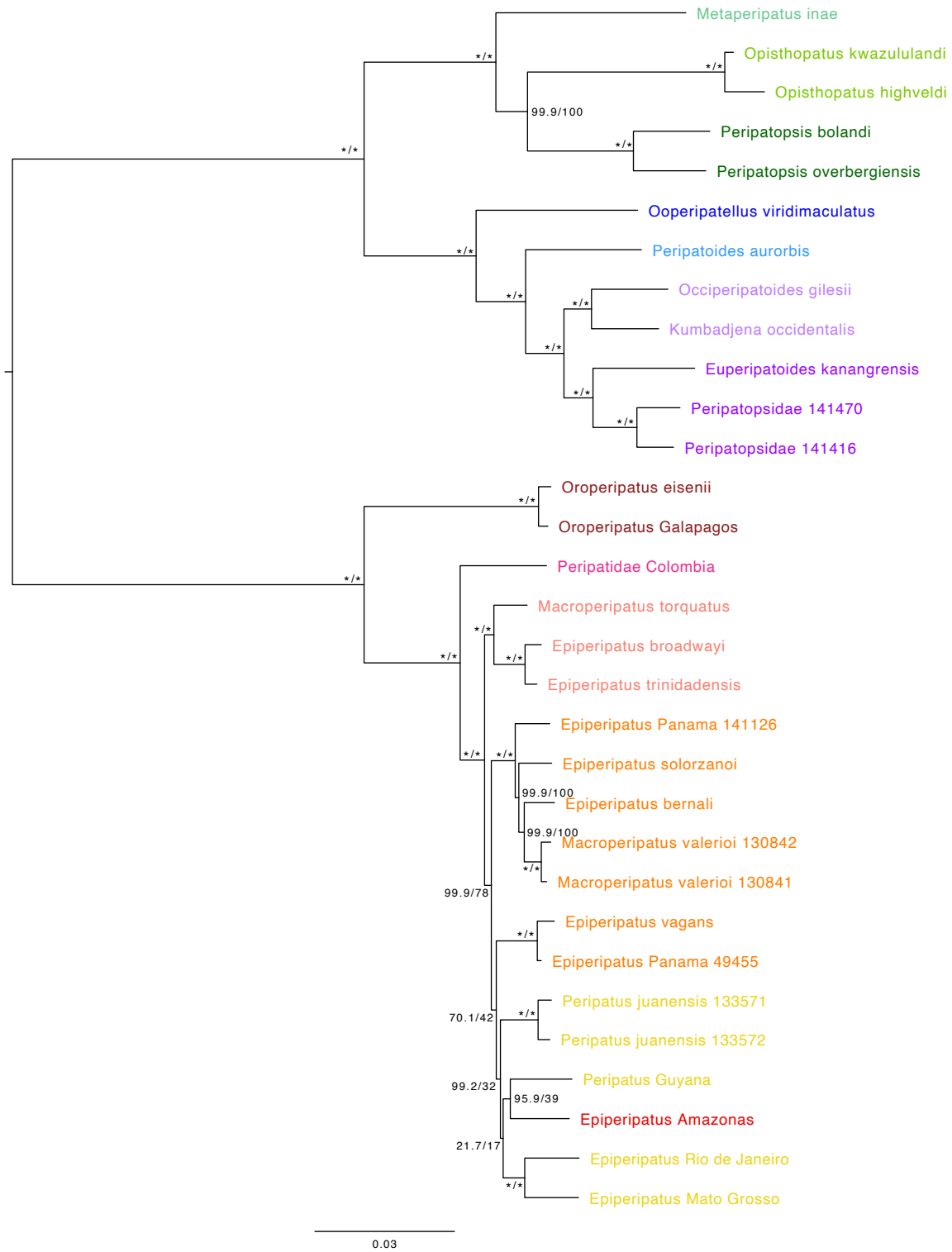


Figure S10. Phylogeny inferred from IQ-TREE analysis of M4. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

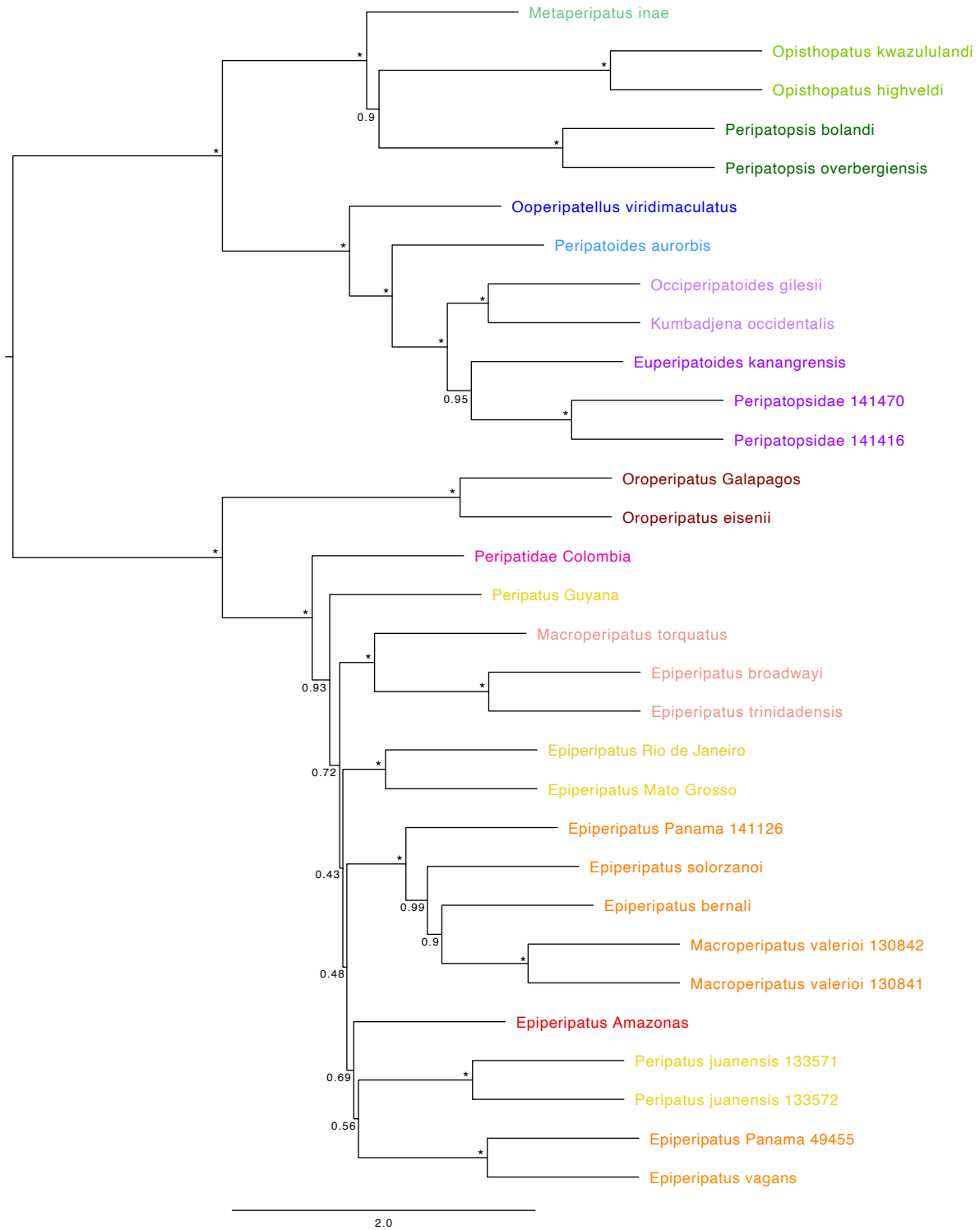


Figure S11. Phylogeny inferred from ASTRAL analysis of M4. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

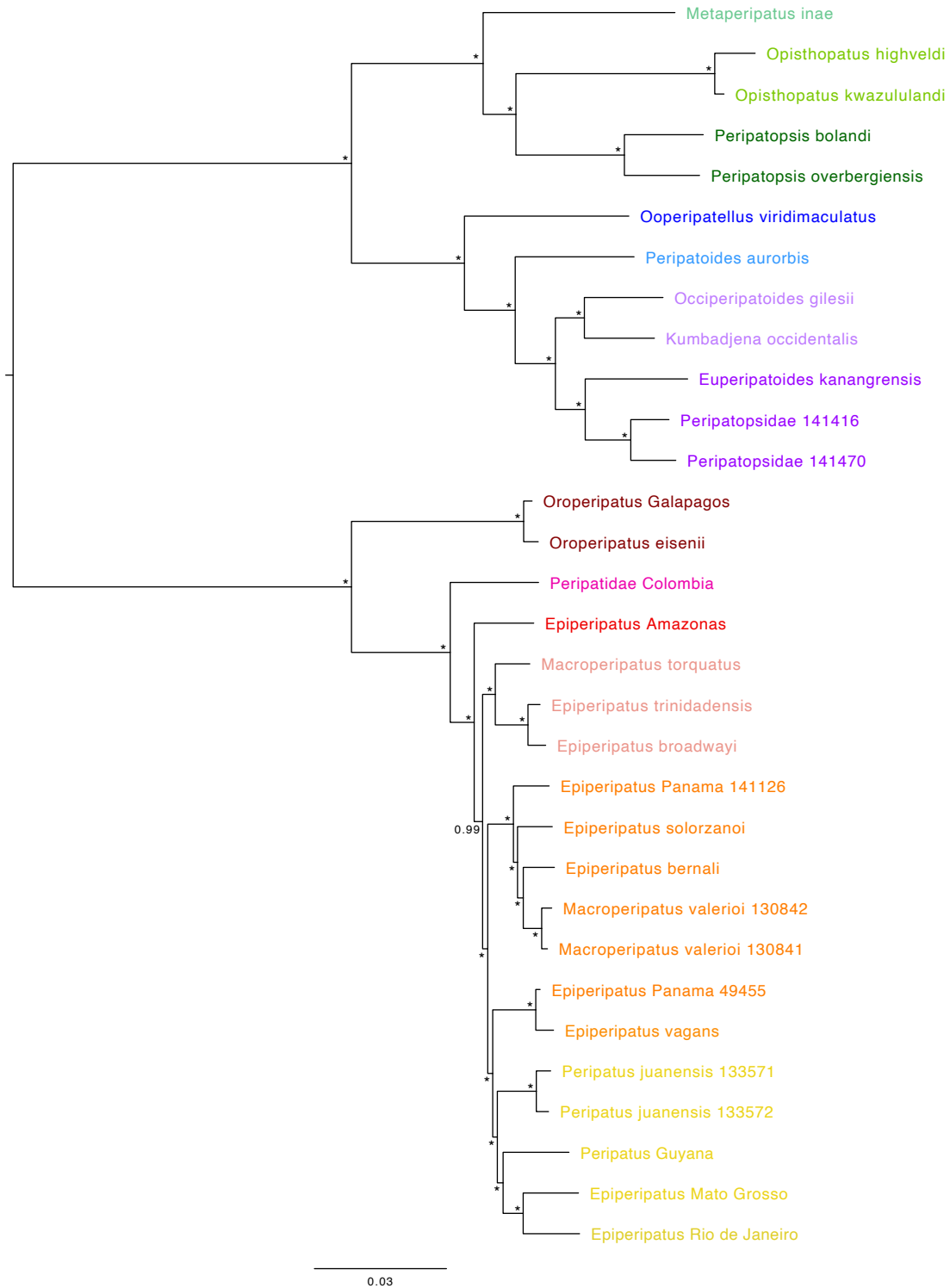


Figure S12. Phylogeny inferred from ExaBayes analysis of M4. Asterisks (*) mark nodes with full support; remaining nodes show the posterior probability.

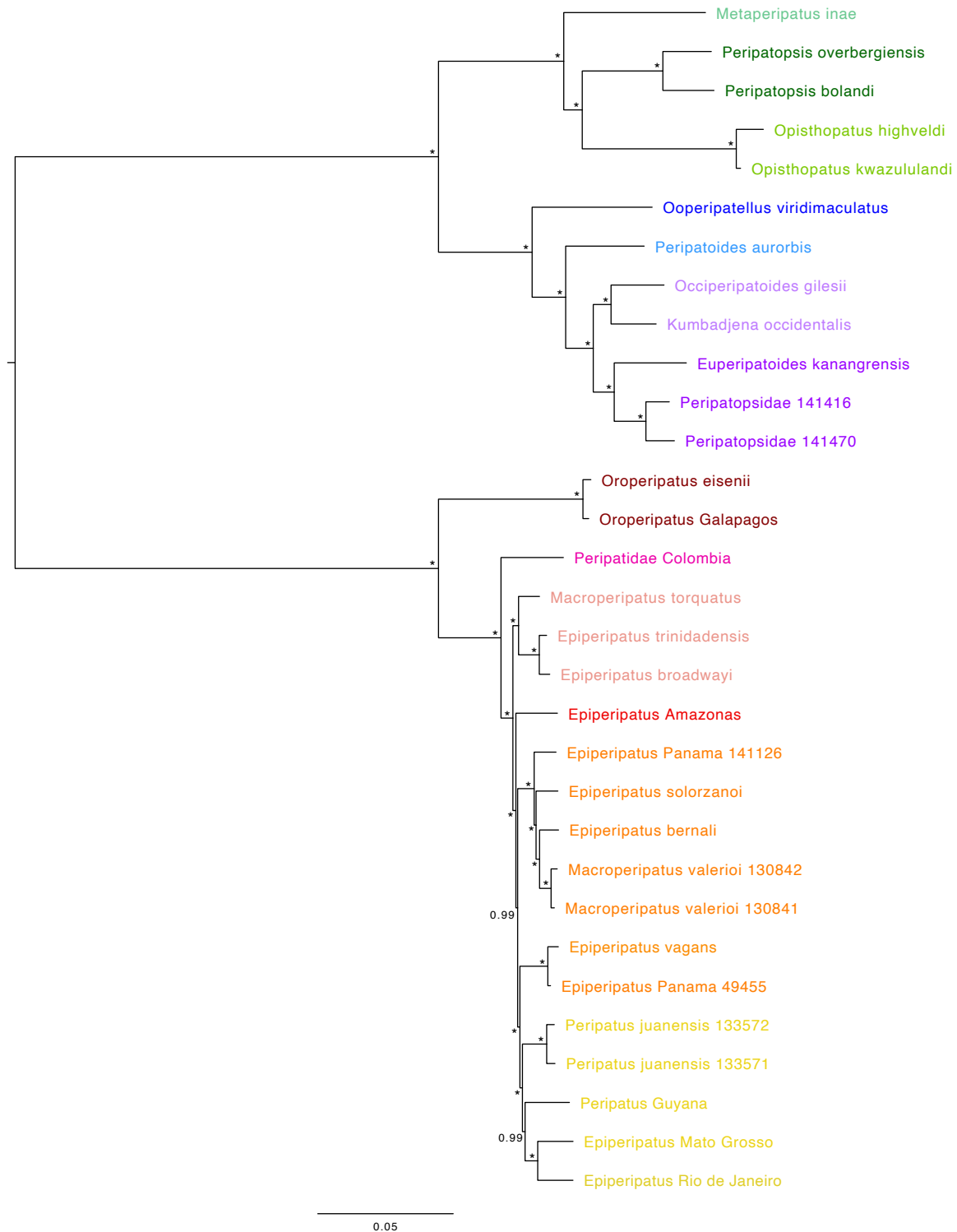


Figure S13. Phylogeny inferred from PhyloBayes analysis of M4. Asterisks (*) mark nodes with full support; remaining nodes show the posterior probability.

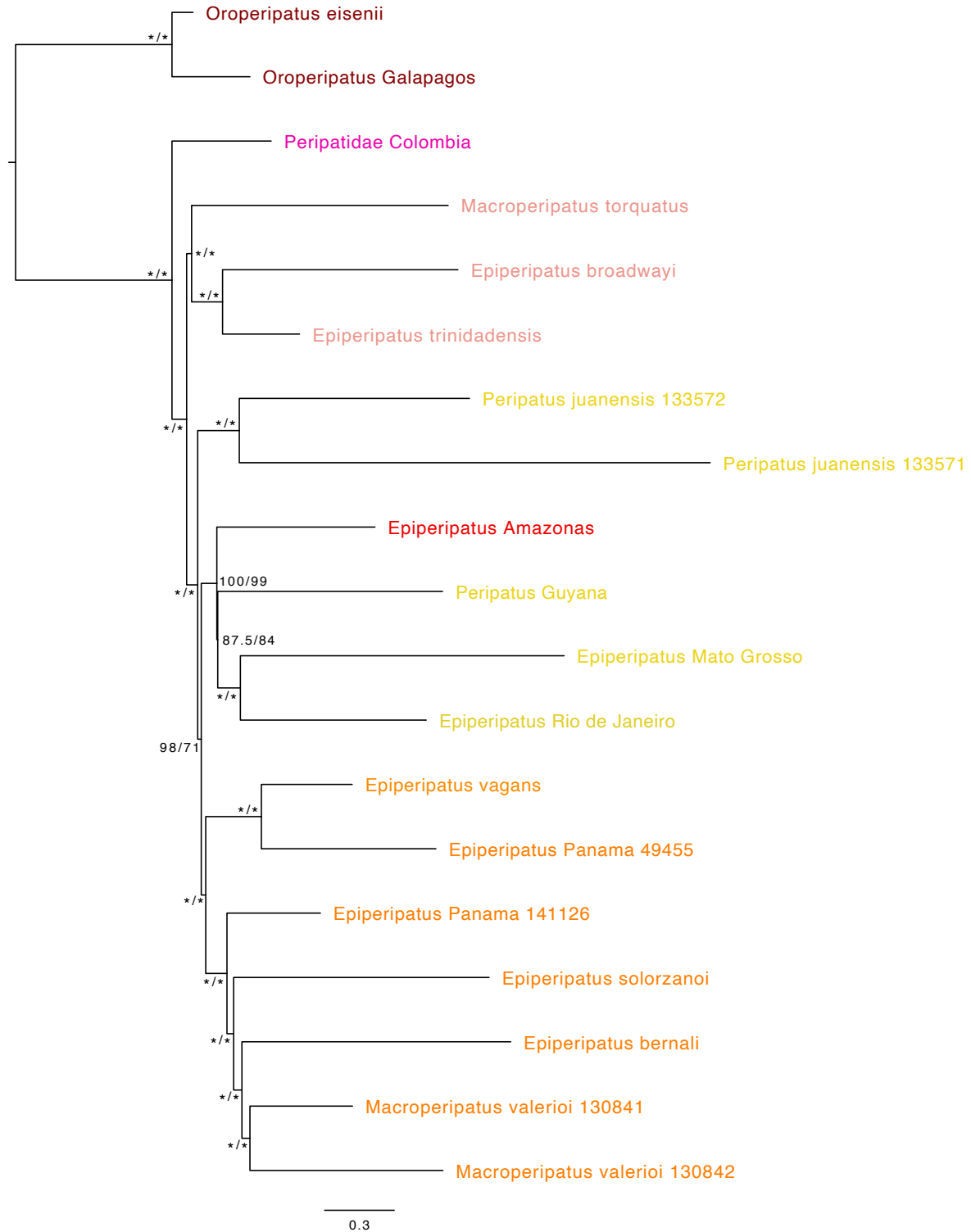


Figure S14. Phylogeny of Neopatida inferred from IQ-TREE analysis of M5. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

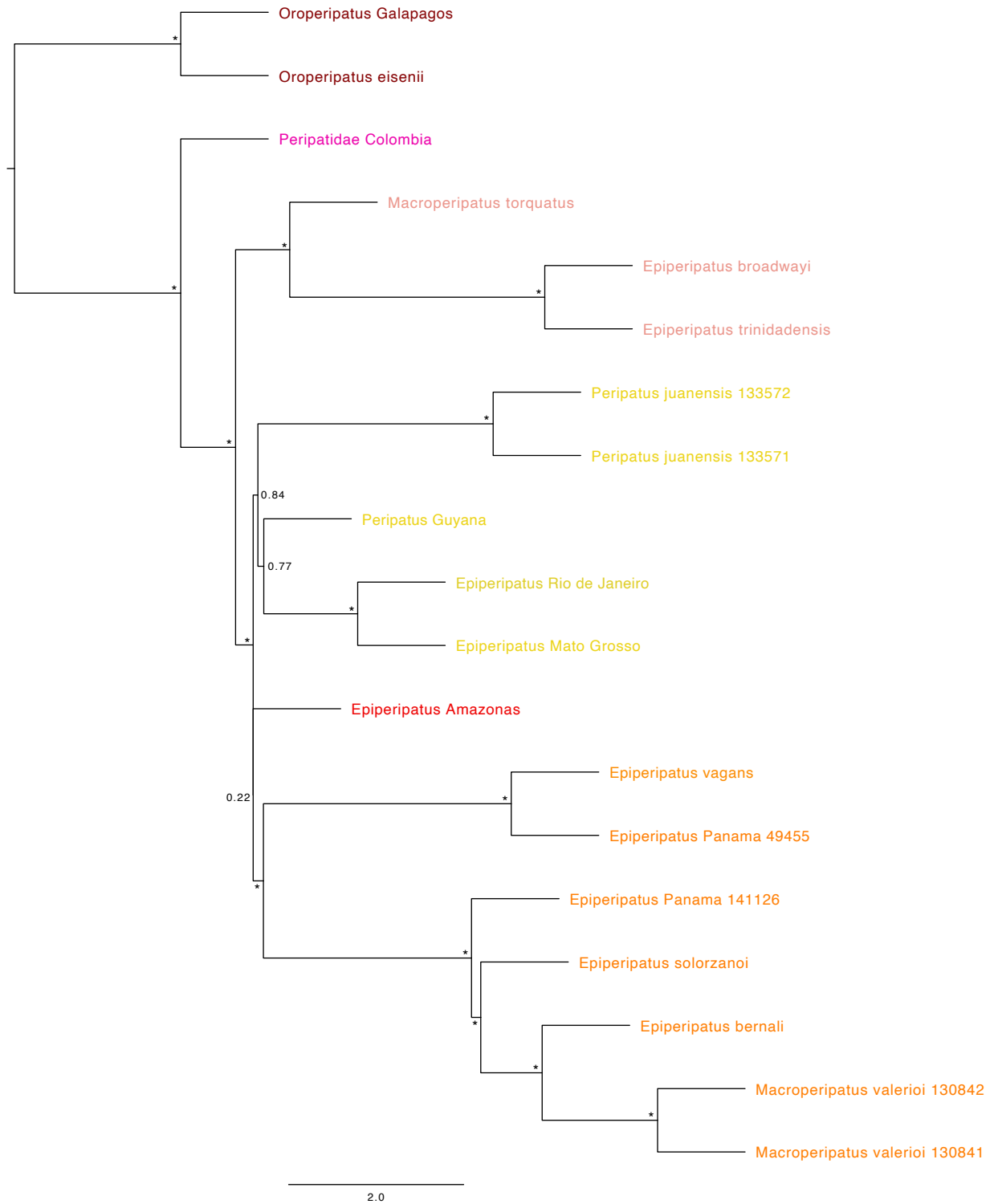


Figure S15. Phylogeny of Neopatida inferred from ASTRAL analysis of M5. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

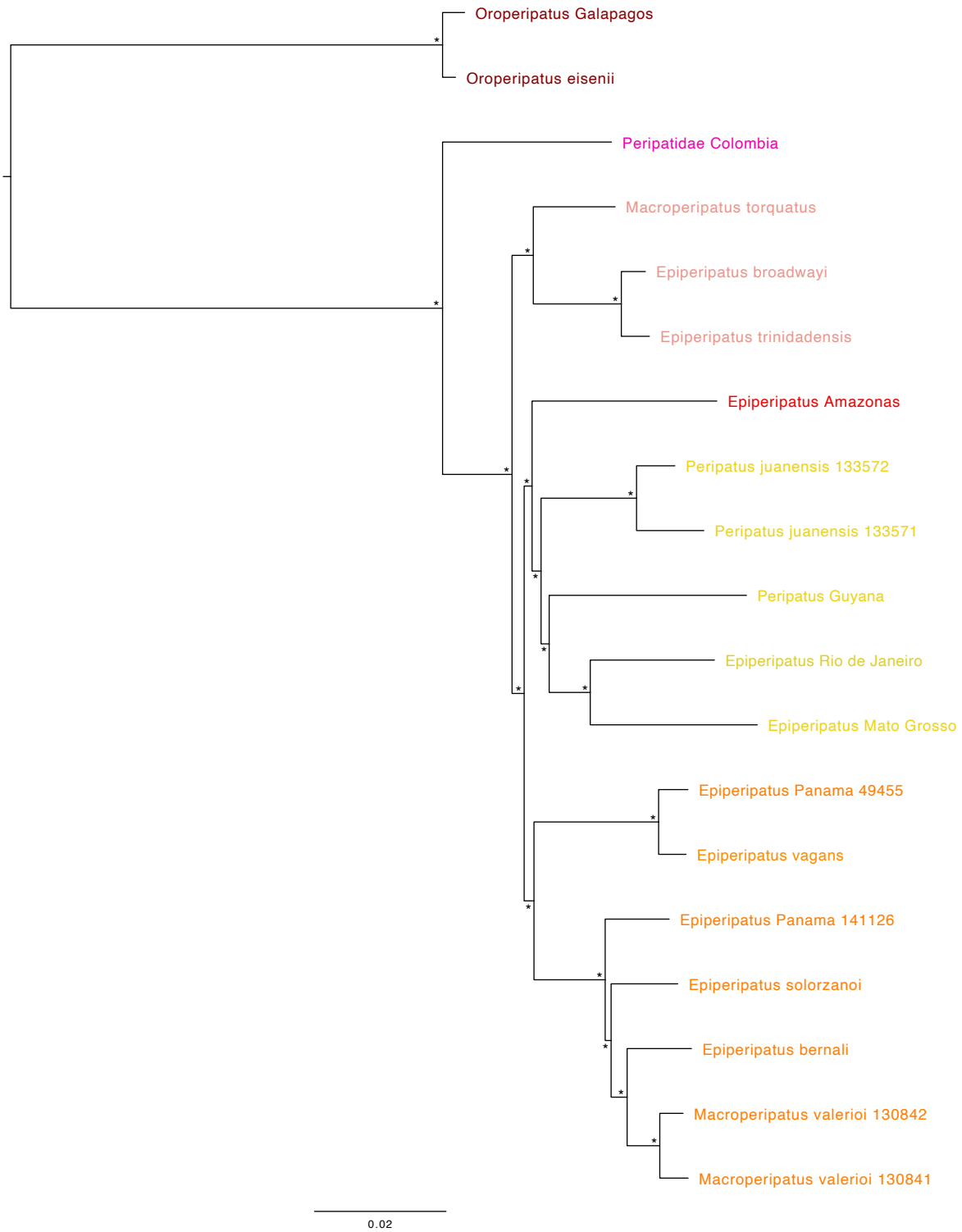


Figure S16. Phylogeny of Neopatida inferred from ExaBayes analysis of M5. Asterisks (*) mark nodes with full support.

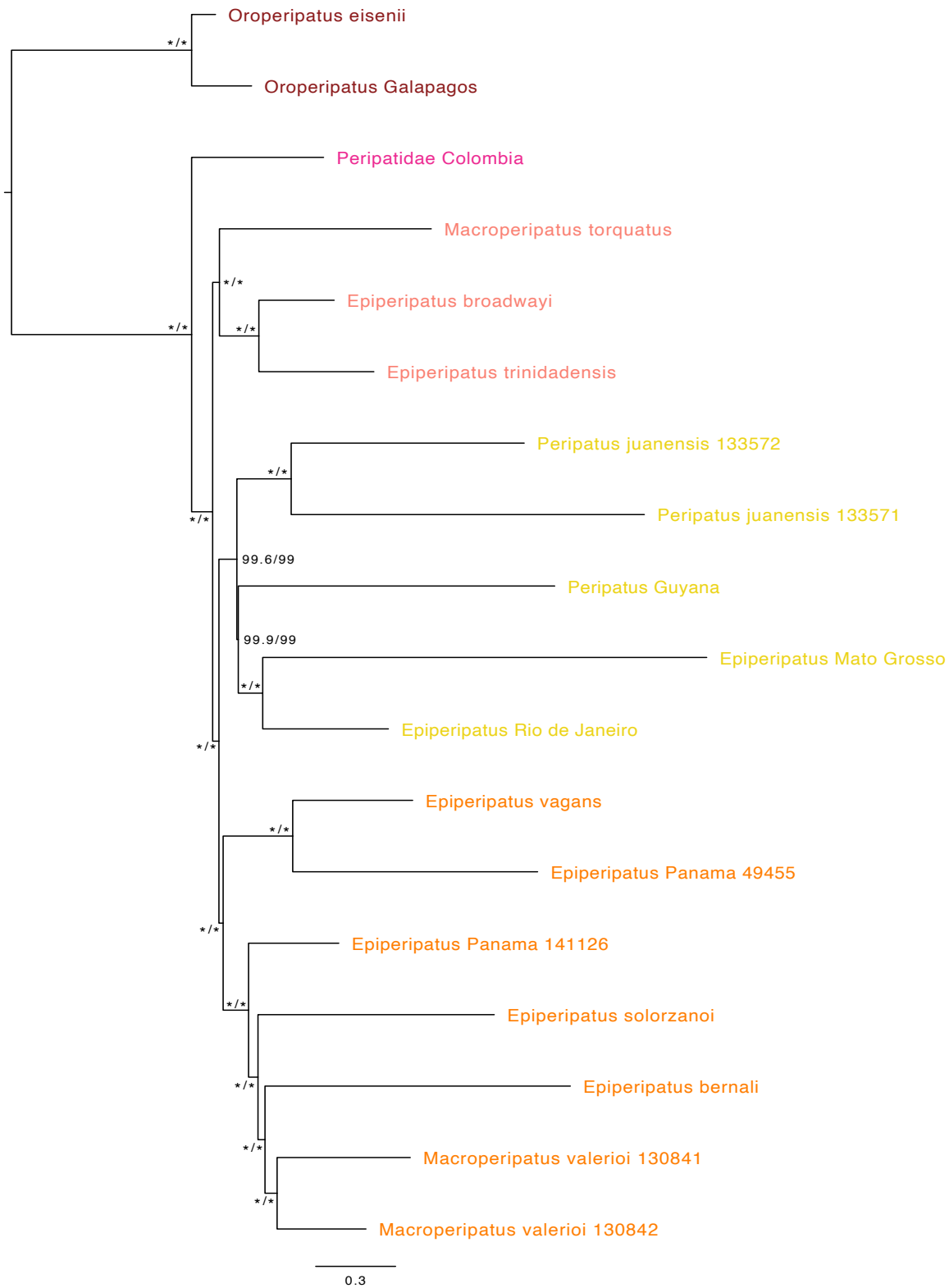


Figure S17. Phylogeny inferred from IQ-TREE analysis of M5, excluding *Epiperipatus* sp. Amazonas (MCZ-136557). Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

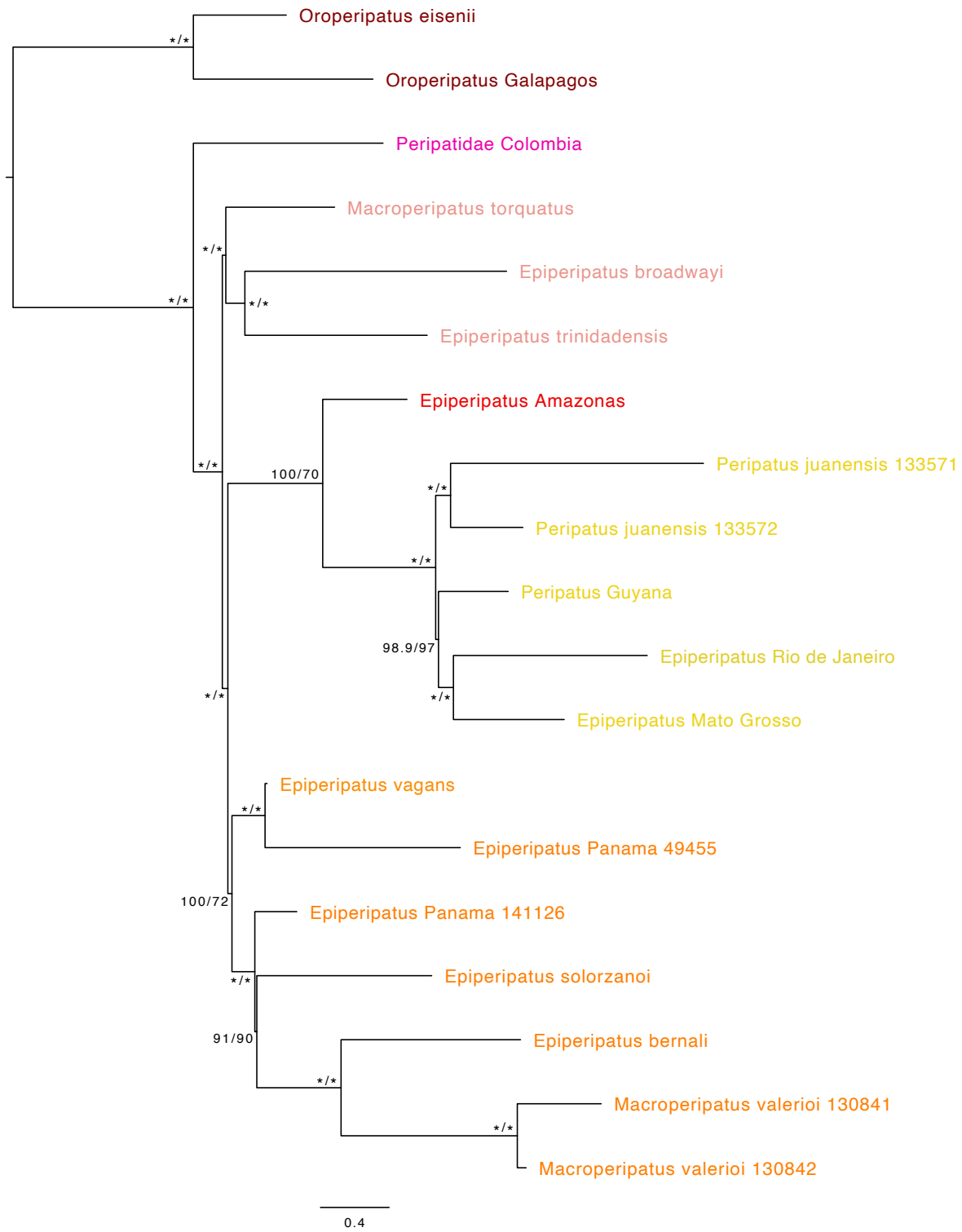


Figure S18. Phylogeny of Neopatida inferred from IQ-TREE analysis of M6. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

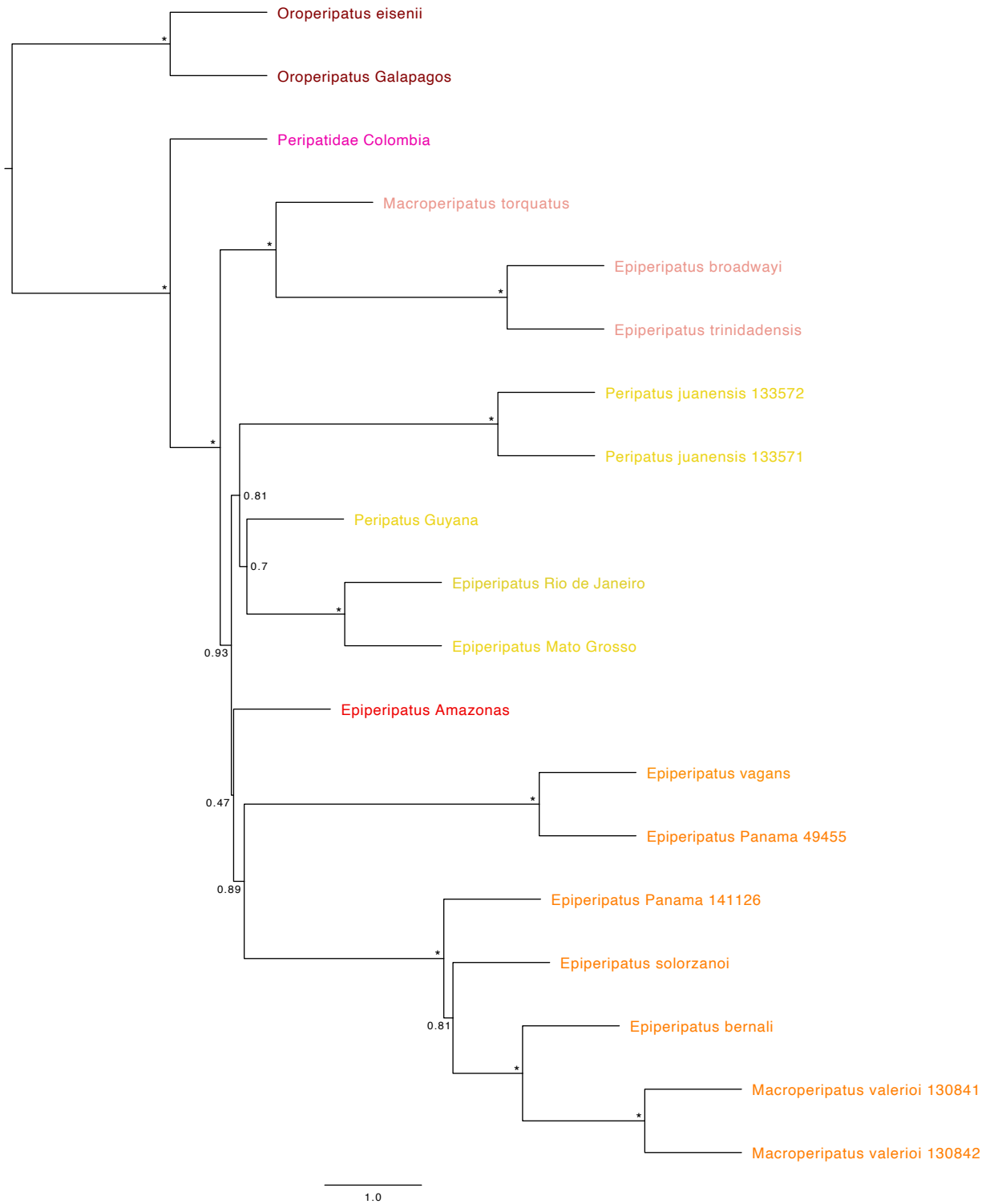


Figure S19. Phylogeny of Neopatida inferred from ASTRAL analysis of M6. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

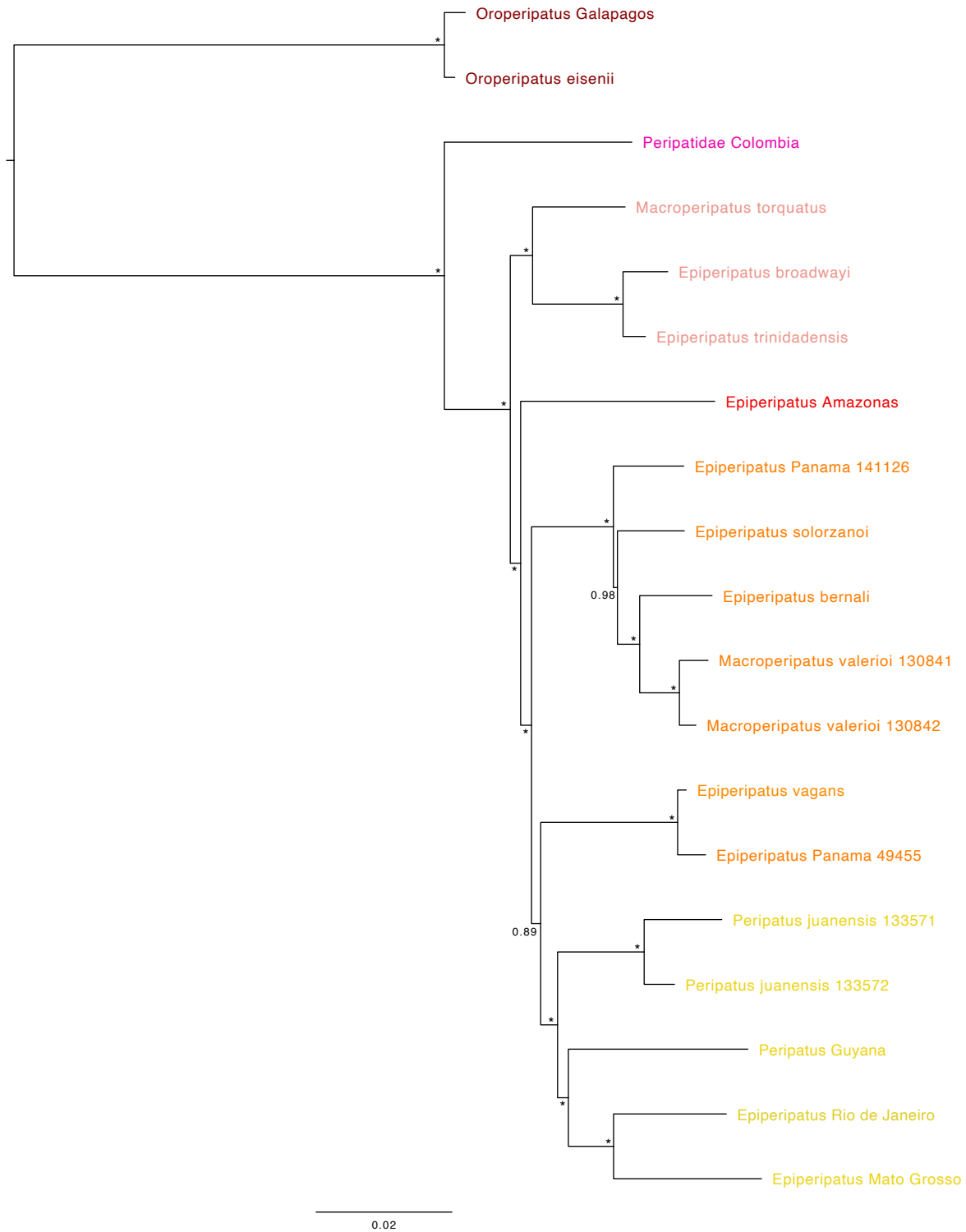


Figure S20. Phylogeny of Neopatida inferred from ExaBayes analysis of M6. Asterisks (*) mark nodes with full support; remaining nodes show the posterior probability.

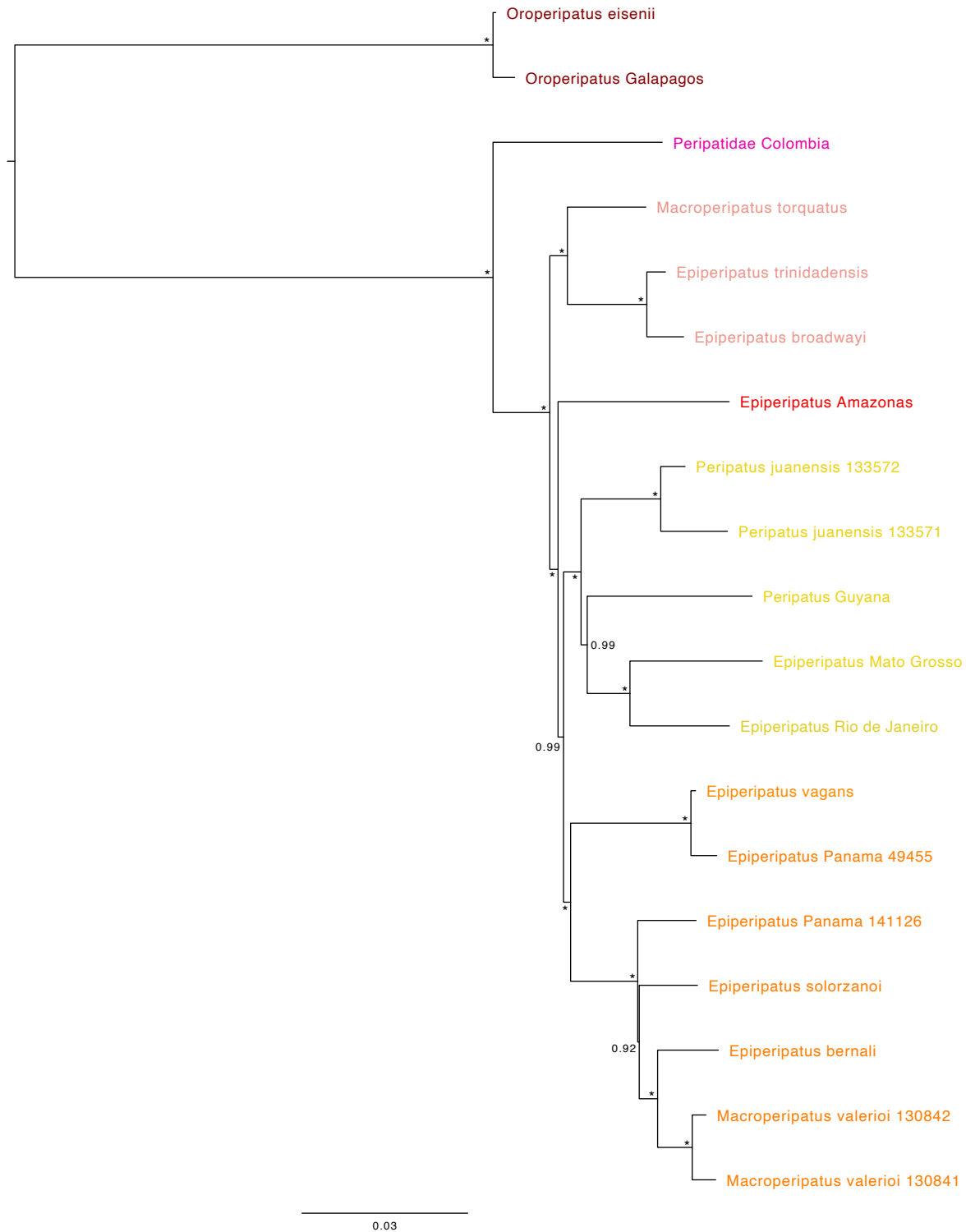


Figure S21. Phylogeny inferred from PhyloBayes analysis of M6. Asterisks (*) mark nodes with full support; remaining nodes show the posterior probability.

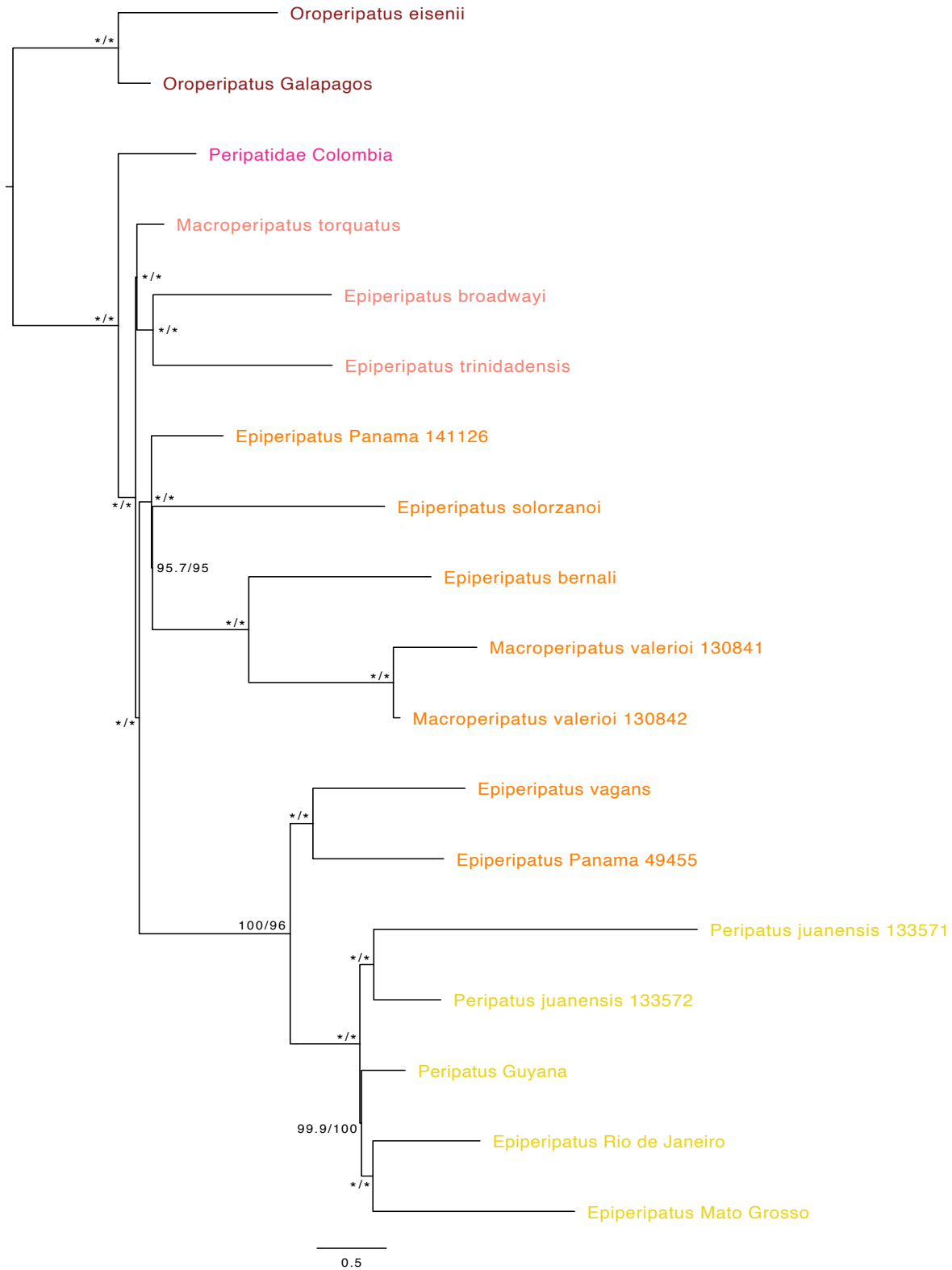


Figure S22. Phylogeny inferred from IQ-TREE analysis of M6, excluding *Epiperipatus* sp. Amazonas (MCZ-136557). Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

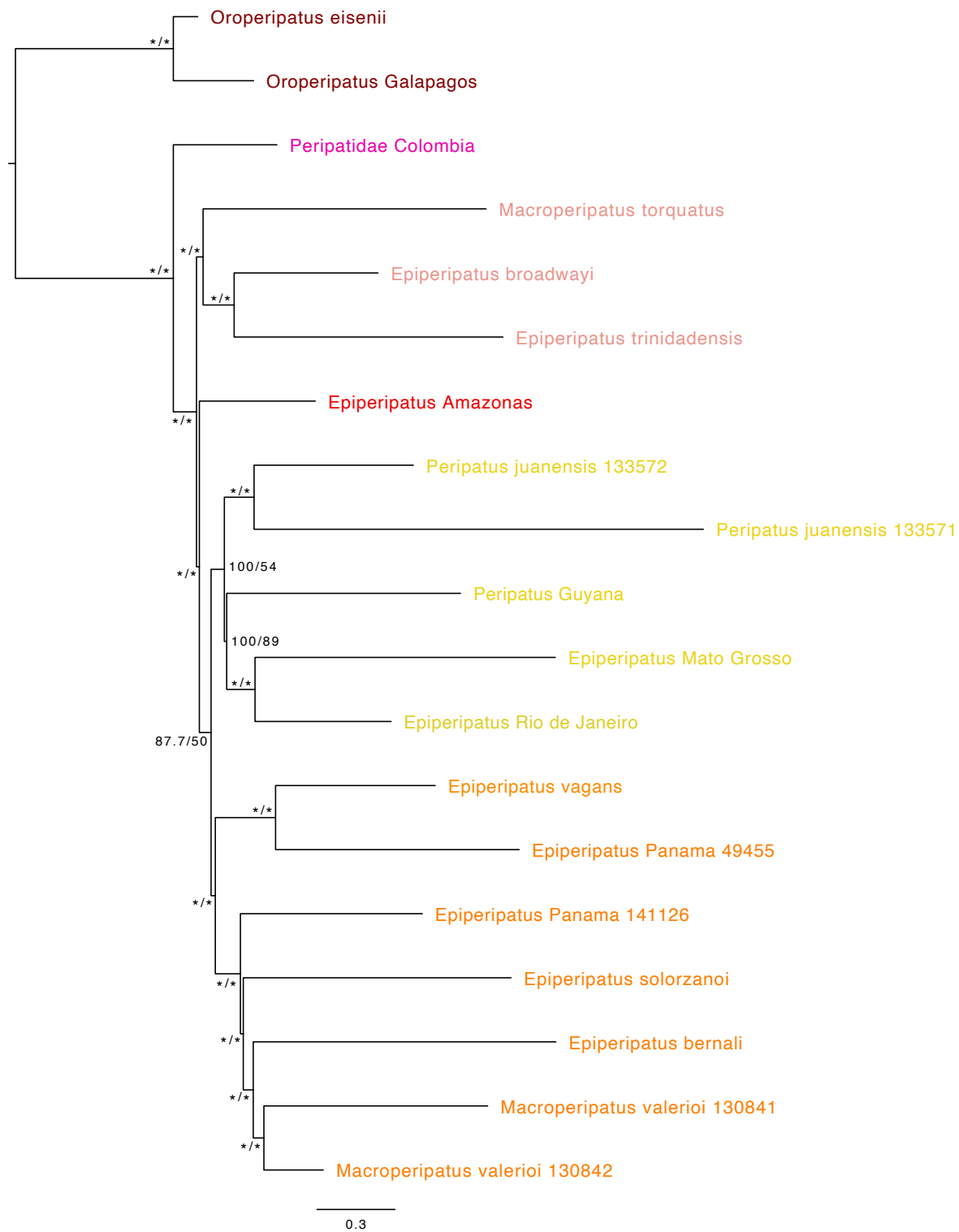


Figure S23. Phylogeny of Neopatida inferred from IQ-TREE analysis of M7. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

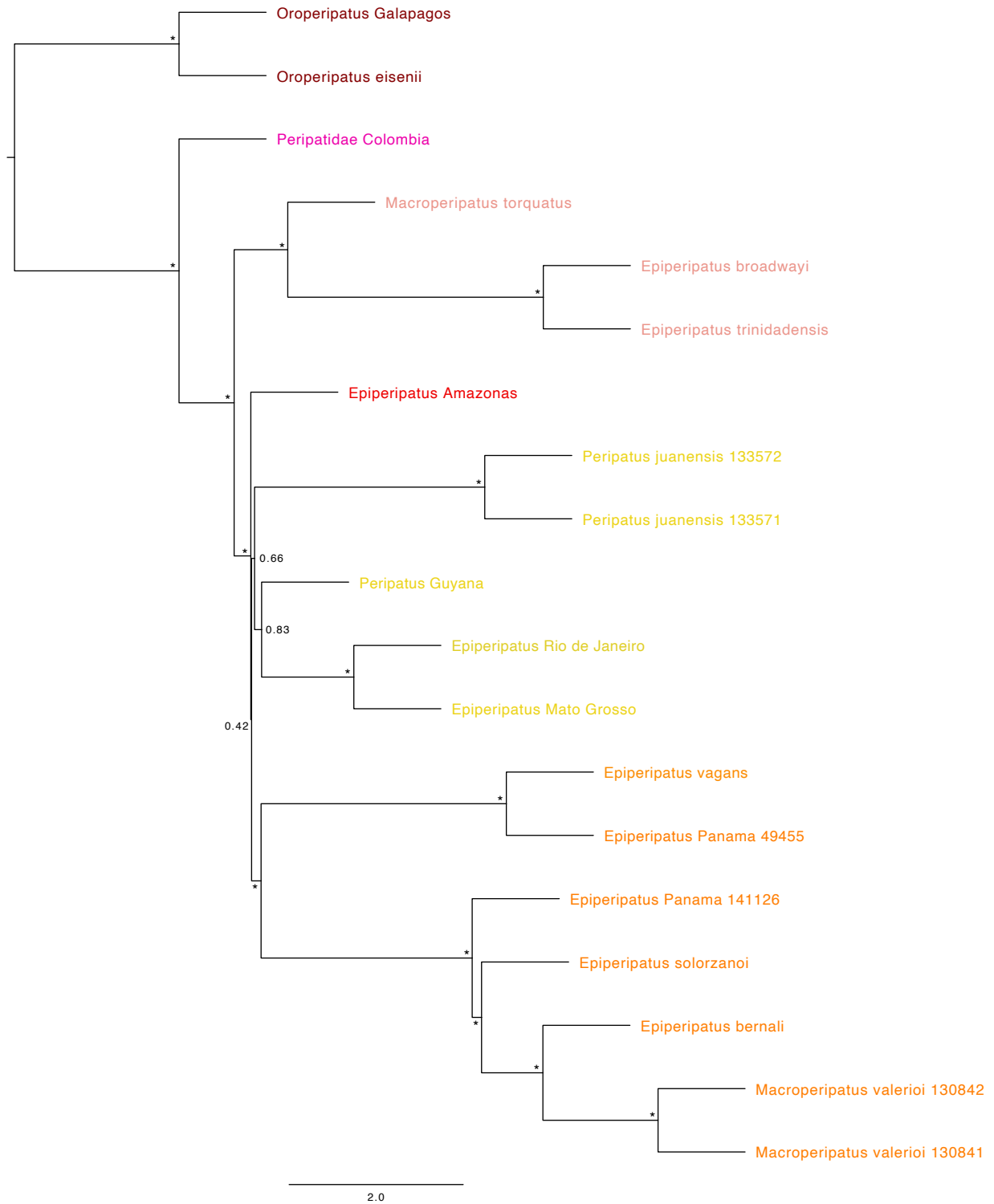


Figure S24. Phylogeny of Neopatida inferred from ASTRAL analysis of M7. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

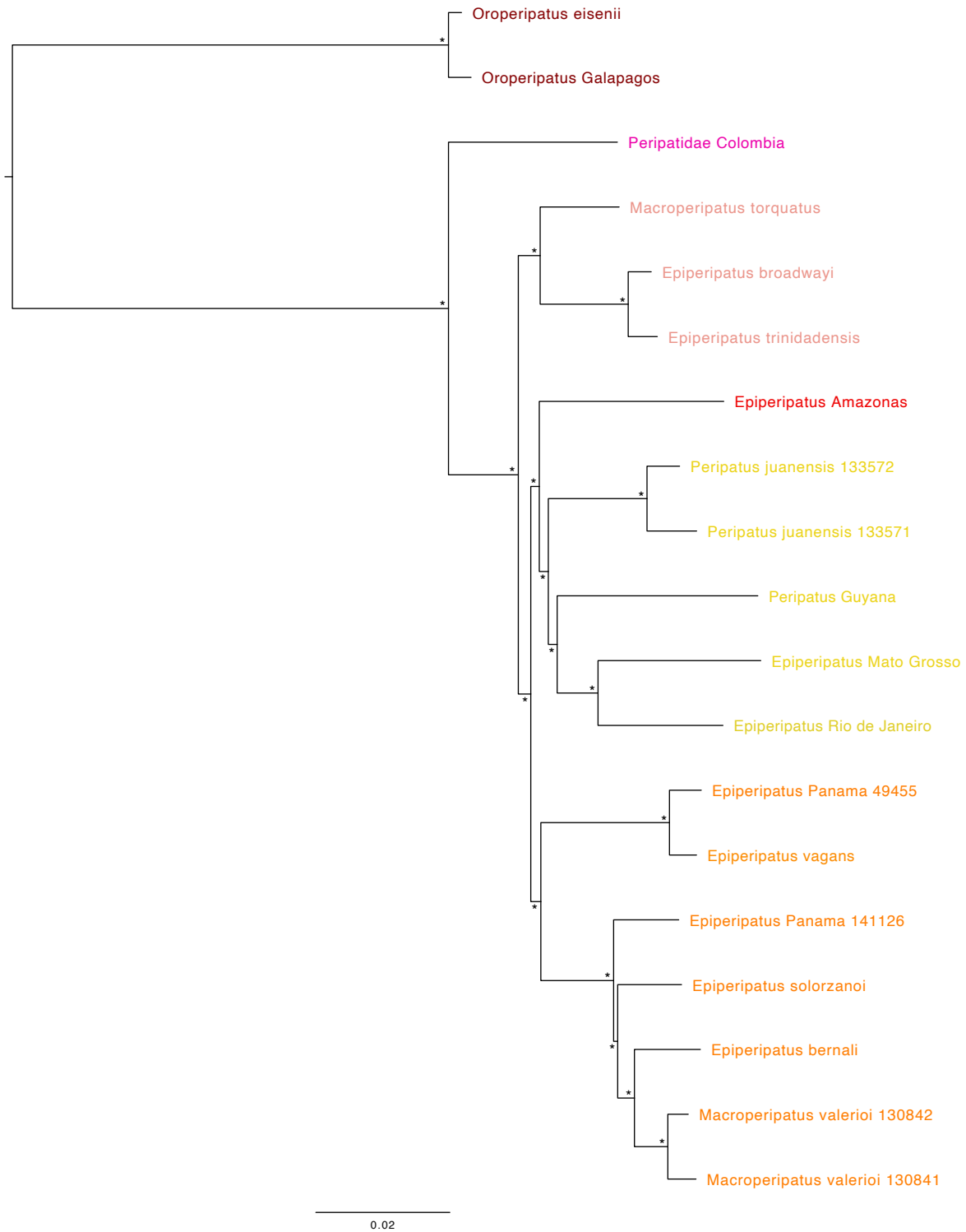


Figure S25. Phylogeny of Neopatida inferred from ExaBayes analysis of M7. Asterisks (*) mark nodes with full support.

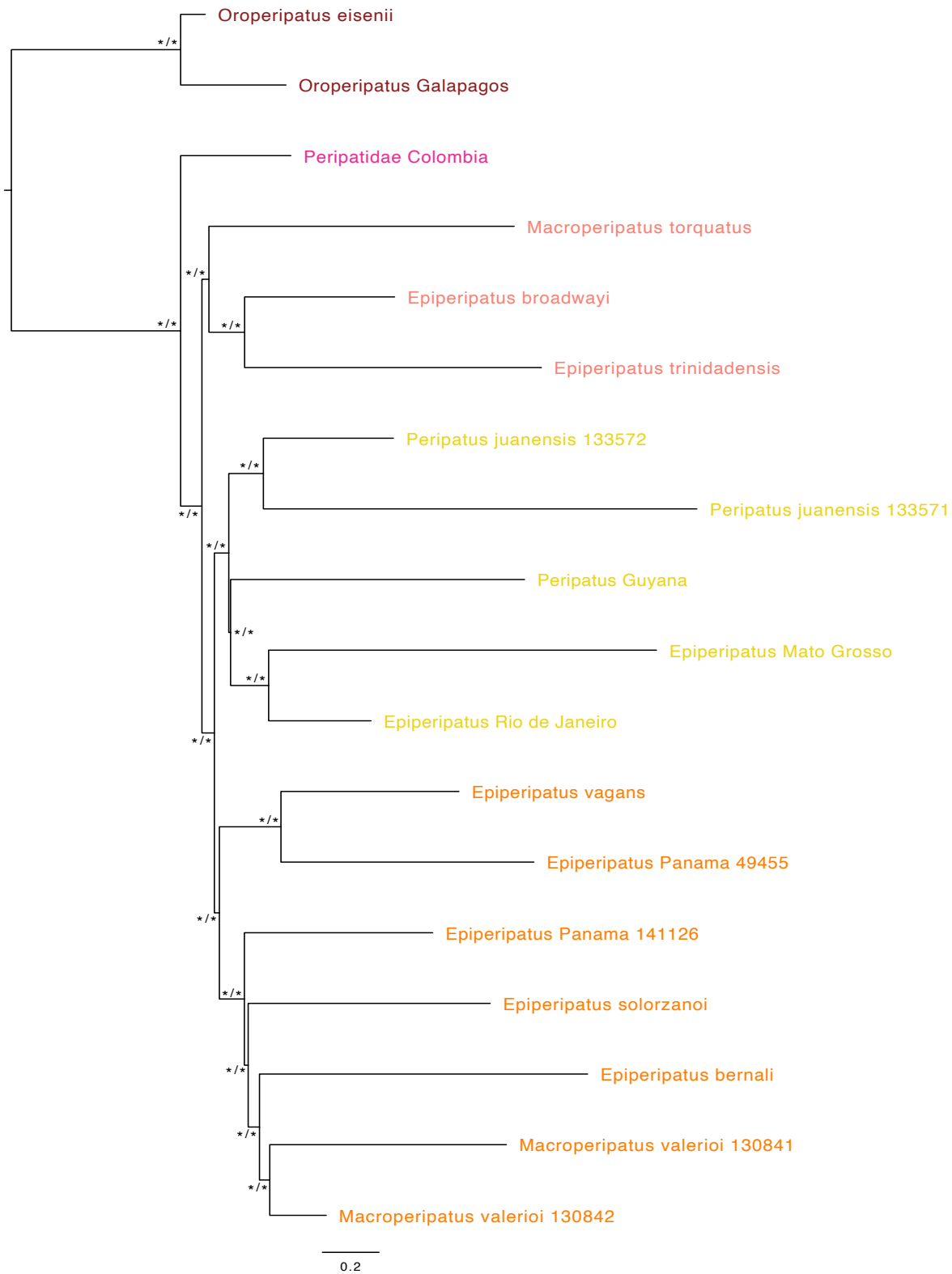


Figure S26. Phylogeny inferred from IQ-TREE analysis of M7, excluding *Epiperipatus* sp. Amazonas (MCZ-136557). Asterisks (*) mark nodes with full support.

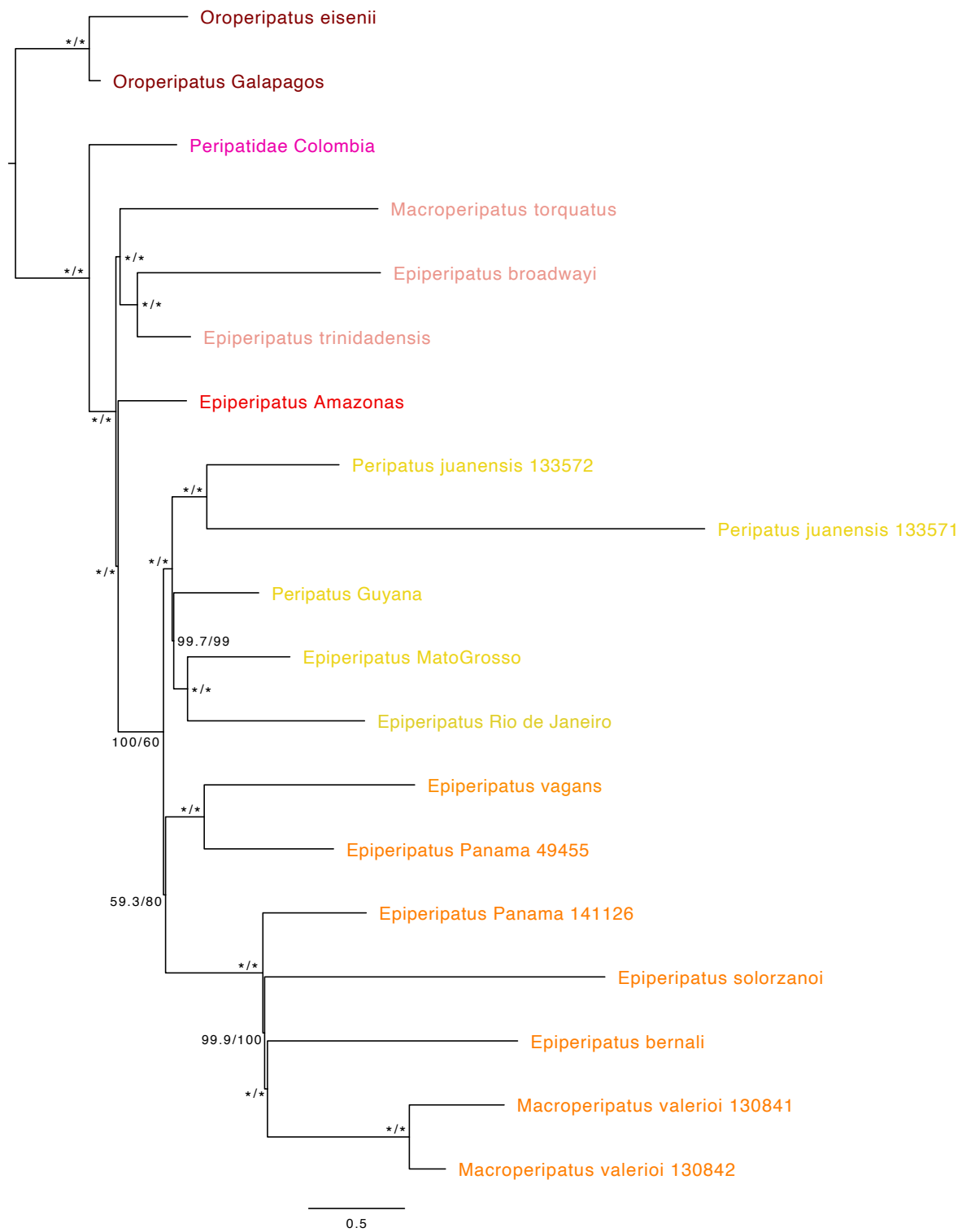


Figure S27. Phylogeny of Neoplatida inferred from IQ-TREE analysis of M8. Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

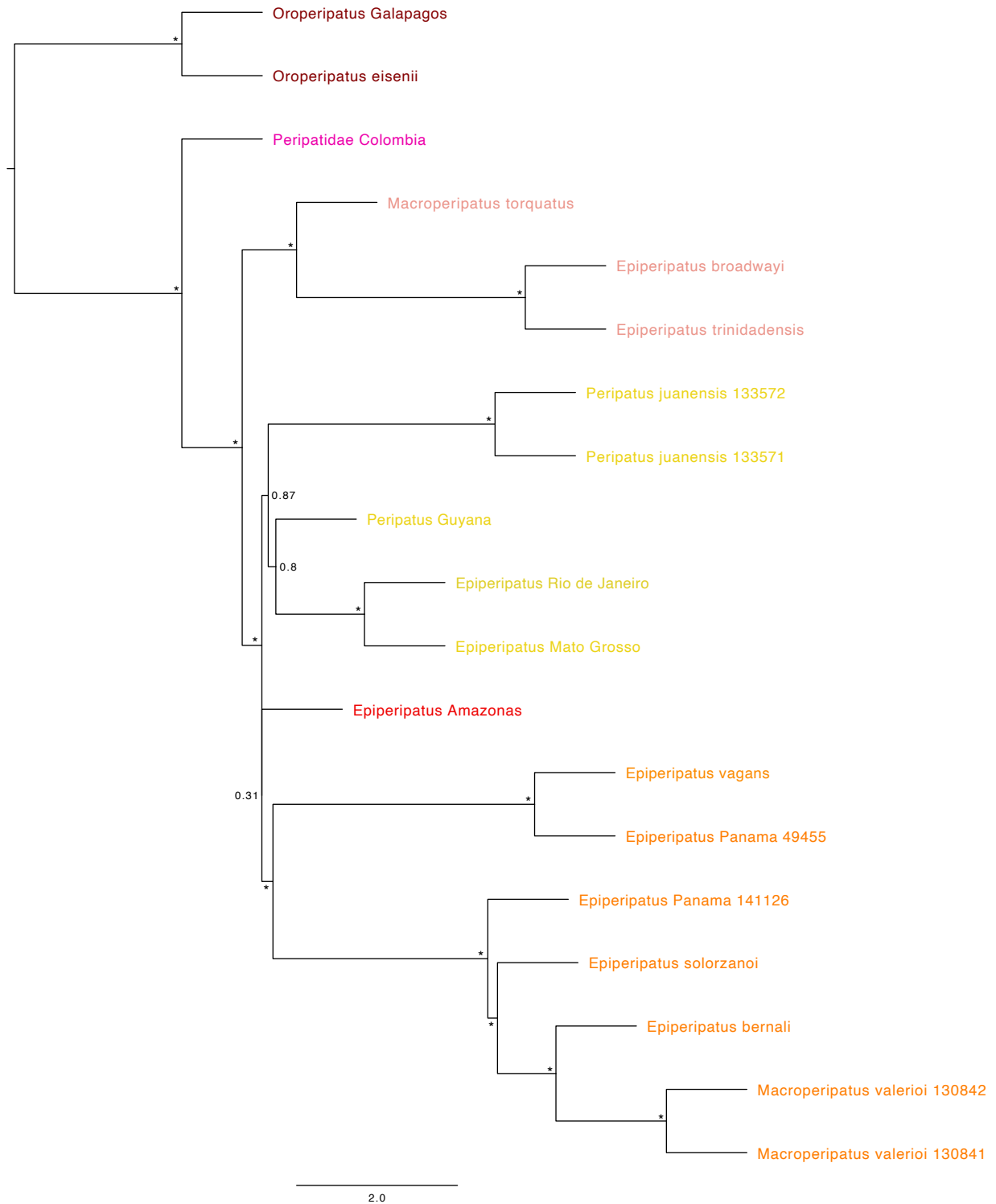


Figure S28. Phylogeny of Neopatida inferred from ASTRAL analysis of M8. Asterisks (*) mark nodes with full support; remaining nodes show local posterior probabilities.

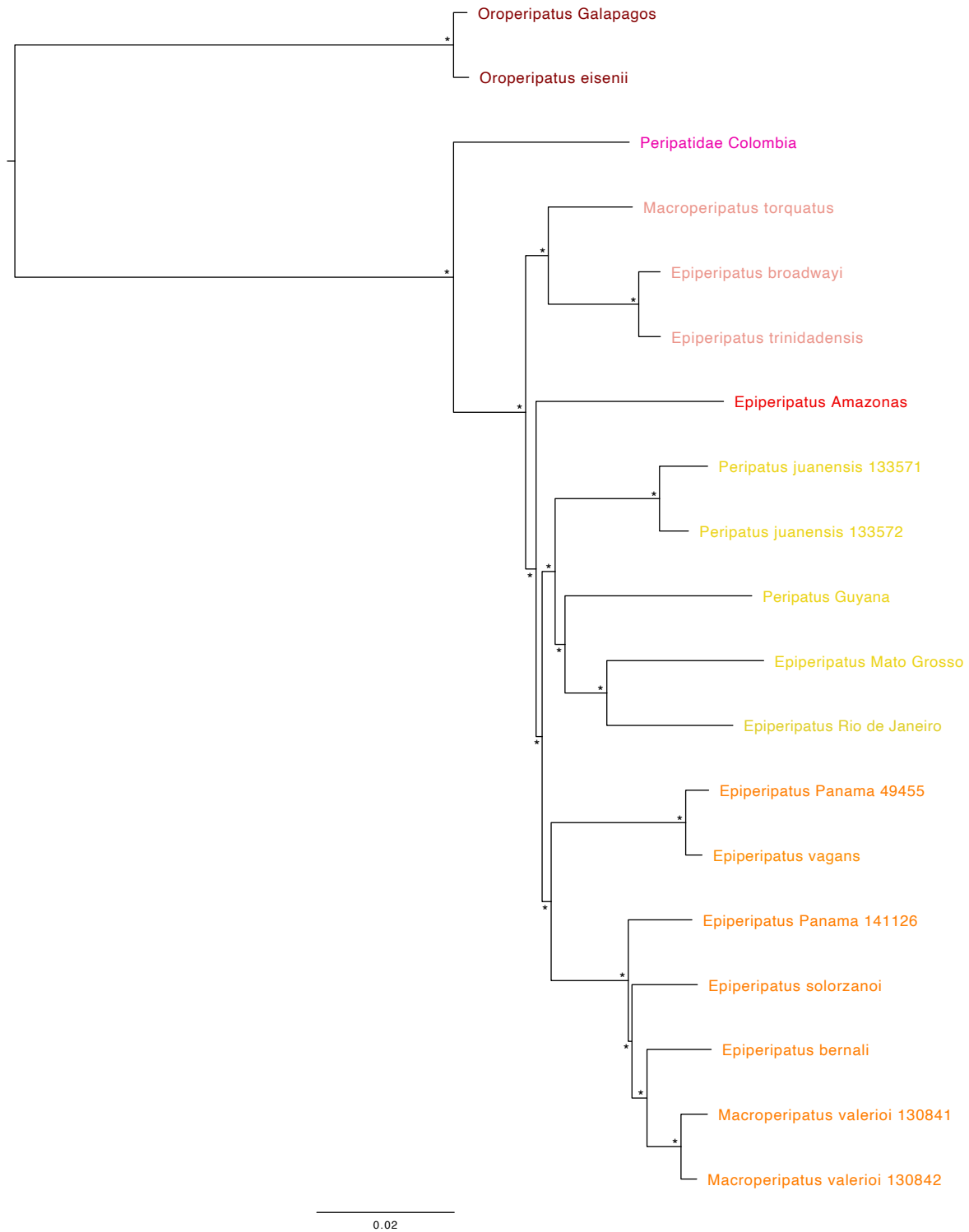


Figure S29. Phylogeny of Neopatida inferred from ExaBayes analysis of M8. Asterisks (*) mark nodes with full support.

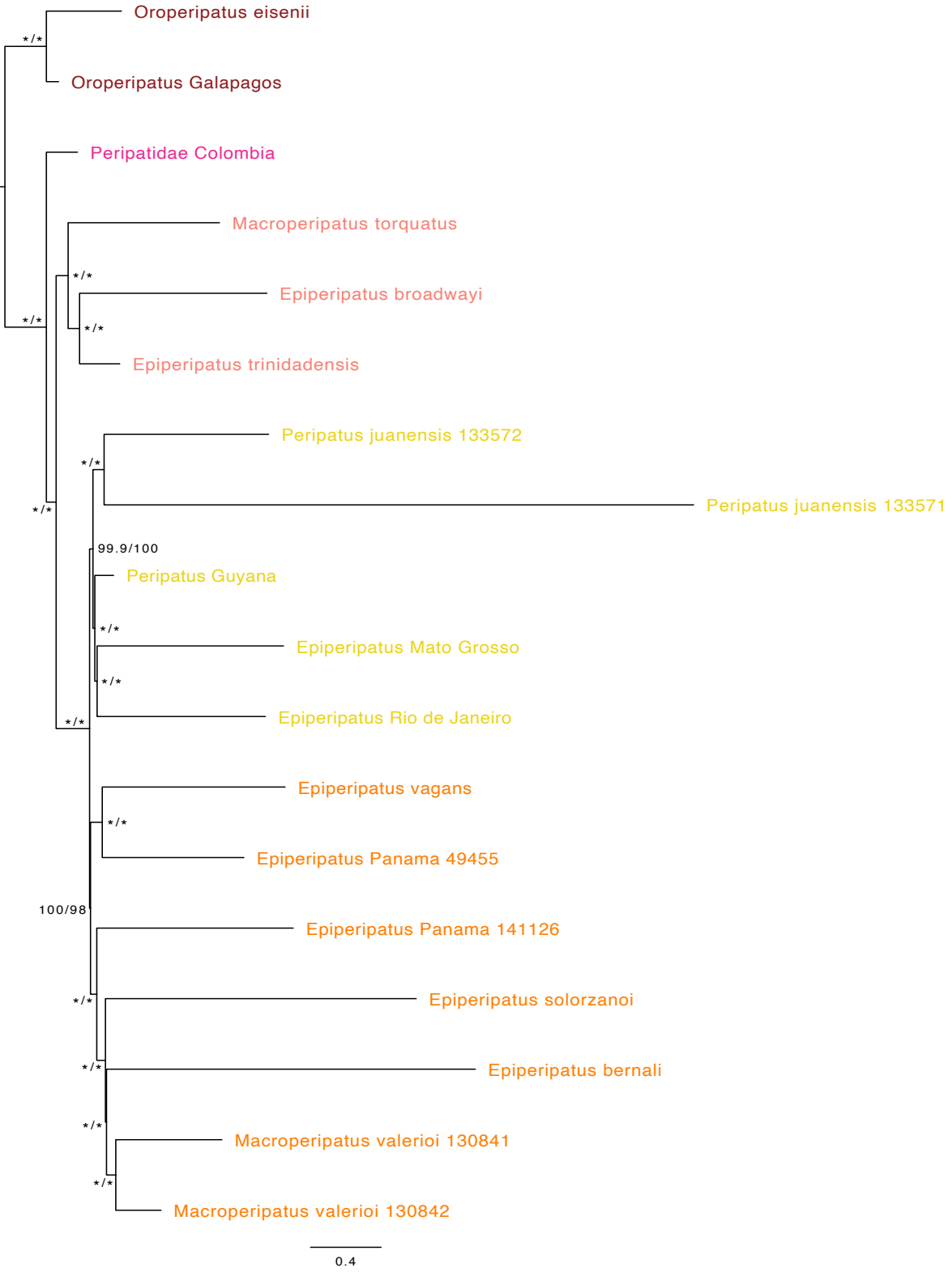


Figure S30. Phylogeny inferred from IQ-TREE analysis of M8, excluding *Epiperipatus* sp. Amazonas (MCZ-136557). Asterisks (*) mark nodes with full support; remaining nodes show the Shimodaira-Hasegawa approximate likelihood ratio test and ultrafast bootstrap support values.

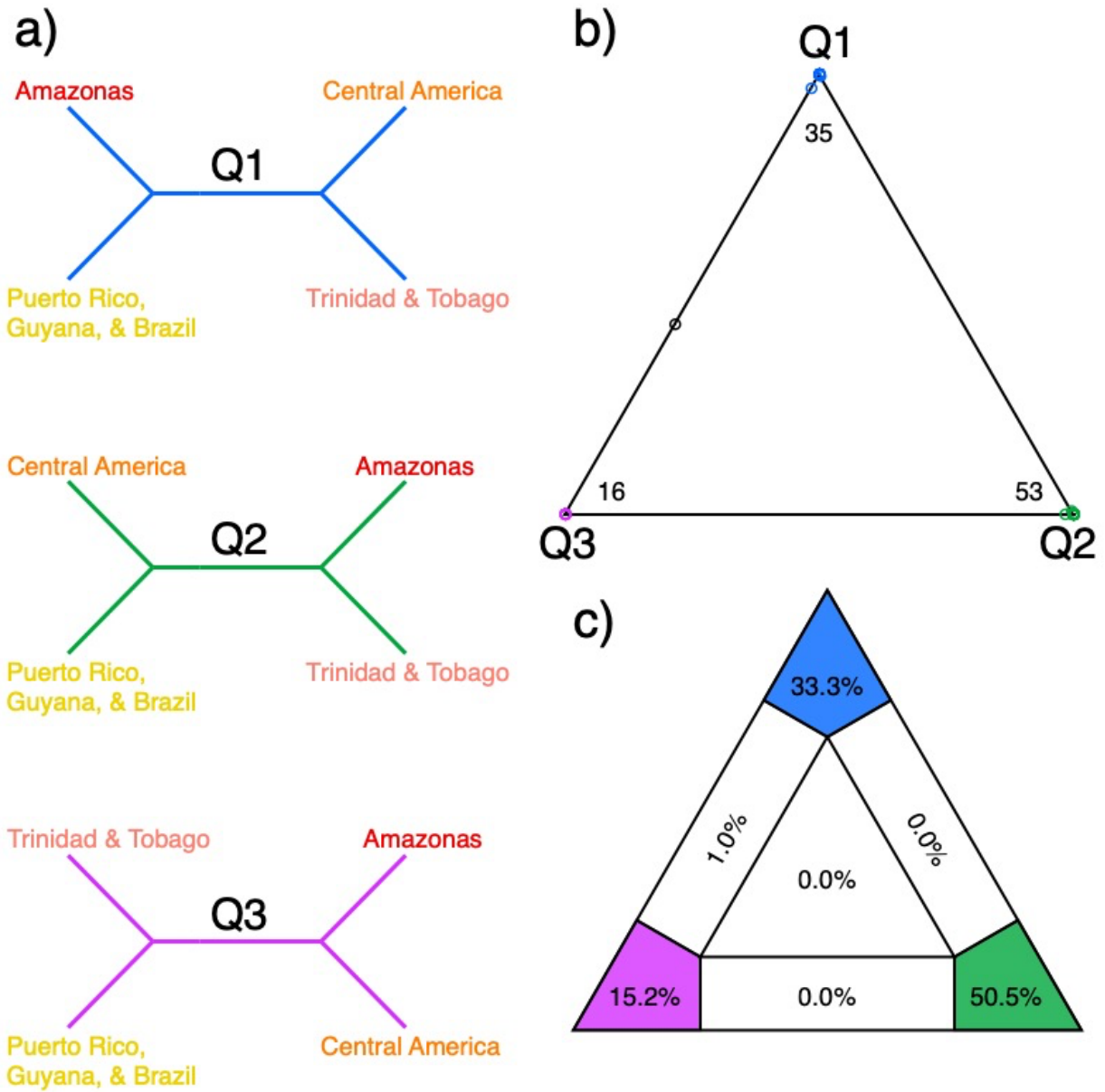


Figure S31. Results of the quartet likelihood mapping analysis performed on M5. (a) The three possible topologies for our four predefined groups from the Neotropics. (b) Number of quartets that support each of the three possible topologies. Points coloured by which topology they support, coloured as in (a). (c) Percentage of quartets that support each of the possible topologies, with regions of the graph differentiated according to how well a quartet falling in that region can answer a phylogenetic hypothesis.

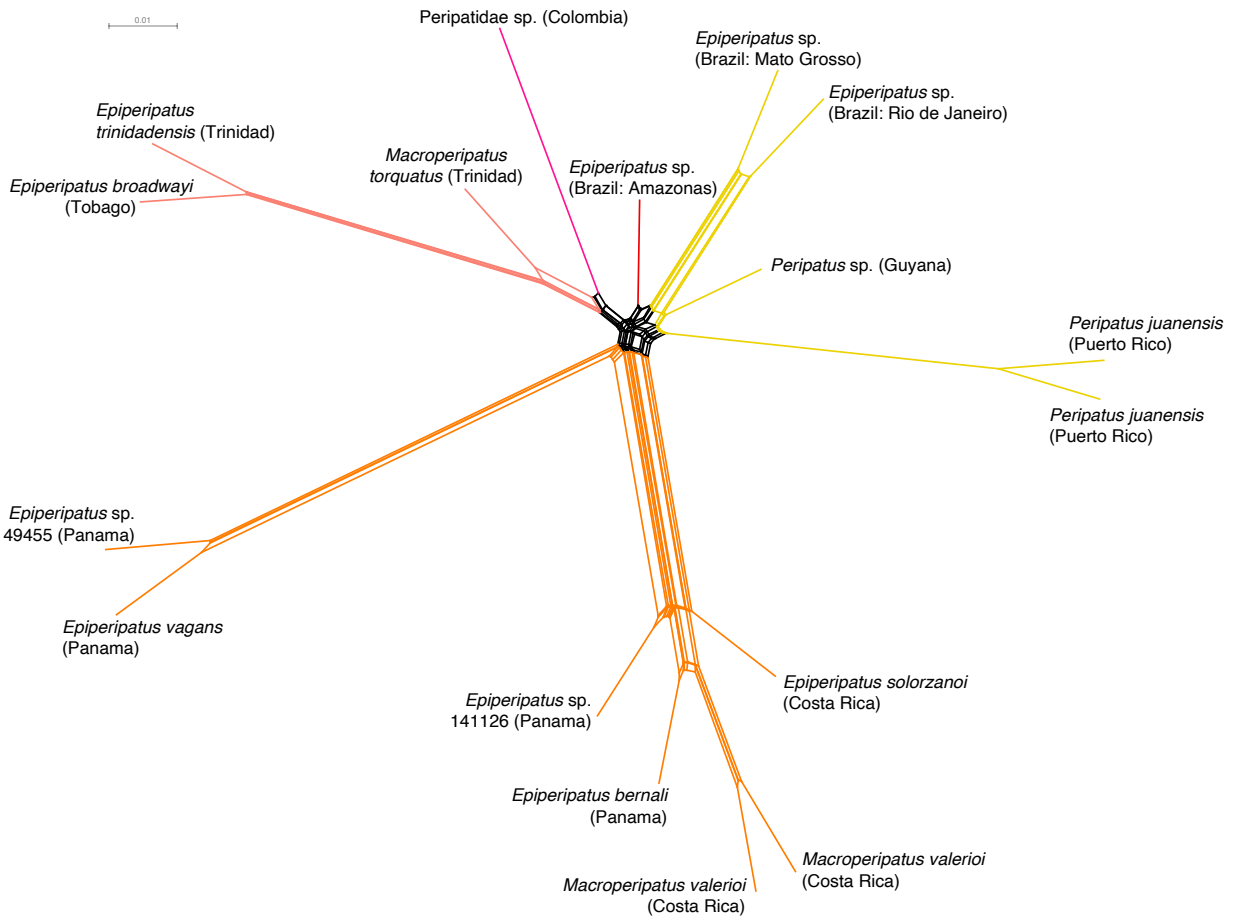


Figure S32. Quartet supernetwork built from 1,147 individual gene trees (M5) in SuperQ, showing conflicting signal between genes. *Oroperipatus eisenii* and *Oroperipatus* sp. Galapagos filtered out for visual clarity. Edges scaled according to bipartition frequency, with specific edges coloured according to clade identity as in main text Figs. 1-3.

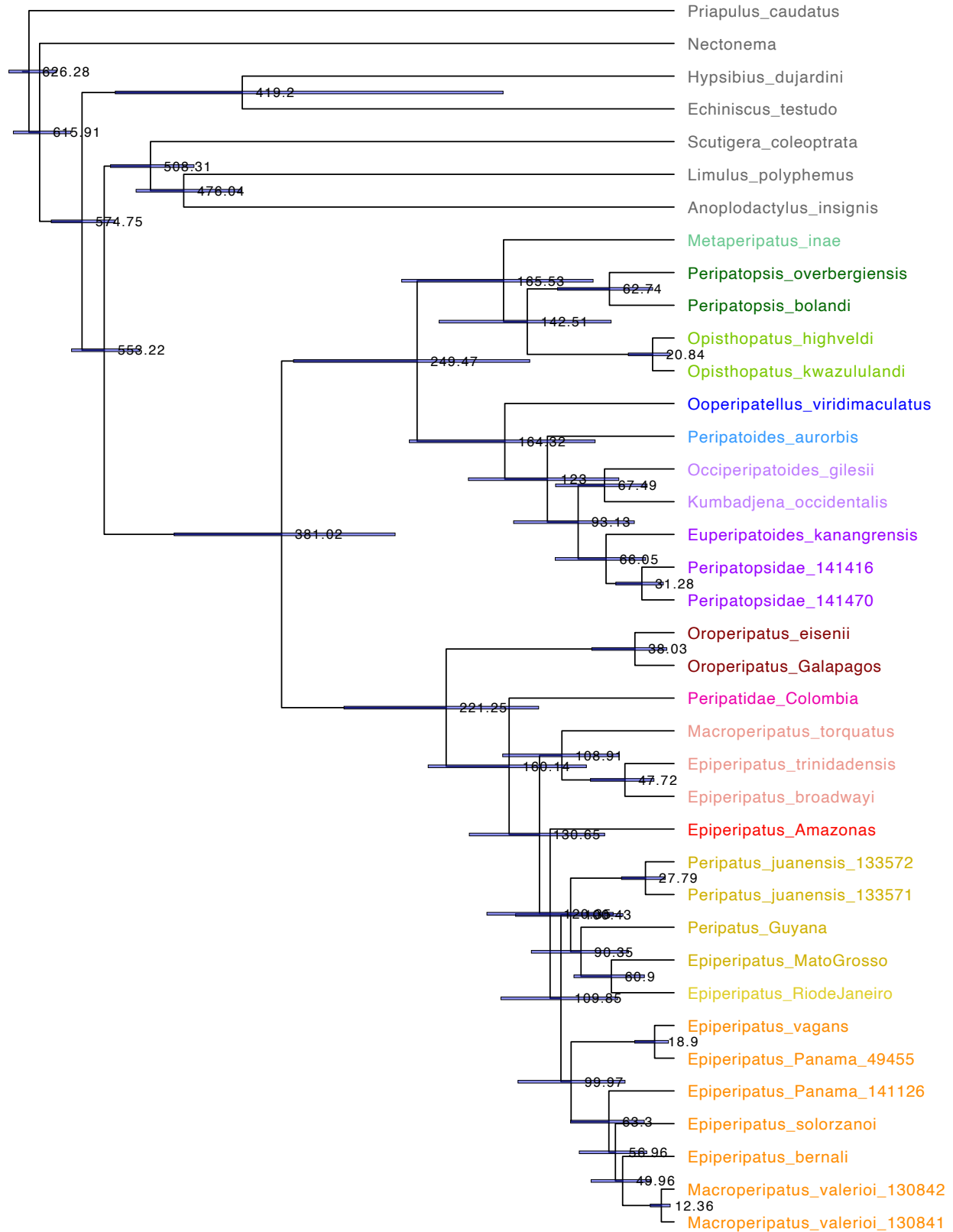


Figure S33. Chronogram inferred in MCMCTree, specifying no constraint on the age of Onychophora. Numbers at nodes show mean node ages, bars show 95% HPD of age estimates.

Table S1. Collection and accession information for onychophoran specimens used in this study. Newly sequenced samples are shown in bold. M3 refers to matrix 3 (see Table S3). BUSCO completeness scores are for the Metazoa database.

Species	MCZ IZ Catalog #	SRA number	Country	BUSCO	Loci in M3	Latitude	Longitude	Collection date	Source
Family Peripatidae									
<i>Epiperipatus</i> sp.	136557	SRR8627713	Brazil (Amazonas)	38.45%	993	-2.9295	-59.9756	18-Dec-2013	(Laumeier et al. 2019)
<i>Epiperipatus</i> sp.	141132	SRR136144 43	Brazil (Mato Grosso)	42.02%	844	-15.4407	-55.7704	23-Dec-2014	this study
<i>Epiperipatus</i> sp.	141131	SRR136144 39	Brazil (Rio de Janeiro)	42.84%	947	-22.4588	-42.6534	21-Dec-2014	this study
Peripatidae sp.	141458	SRR136144 44	Colombia	68.61%	1016	5.8806	-73.0640	4-Dec-2016	this study
<i>Macroperipatus valerioi</i>	130841	SRR136144 28	Costa Rica	39.78%	962	9.4885	-83.9568	13-Jul-2015	this study
<i>Macroperipatus valerioi</i>	130842	SRR136144 27	Costa Rica	14.83%	436	9.4838	-83.9396	8-Jul-2015	this study
<i>Epiperipatus solorzanoi</i>	130840	SRR136144 34	Costa Rica	40.18%	829	9.6731	-83.0243	14-Jun-2015	this study
<i>Oroperipatus</i> sp.	133614	SRR136144 45	Galapagos	9.82%	410	-0.6249	-90.3855	Feb-2016	this study
<i>Peripatus</i> sp.	46445	SRR136144 37	Guyana	10.53%	617	4.1514	-58.2146	22-Sep-2014	this study
<i>Oroperipatus eisenii</i>	74293	SRR136144 46	Mexico	46.63%	836	21.4794	-105.0776	Nov-2015	this study
<i>Epiperipatus</i> sp.	49455	SRR136144 25	Panama	14.01%	725	8.7916	-78.4524	19-Jan-2015	this study
<i>Epiperipatus</i> sp.	141126	SRR8320992	Panama	77.61%	936	8.7467	-82.4189	9-Sep-2014	(Mapaló et al. 2020)
<i>Epiperipatus vagans</i>	141130	SRR136144 32	Panama	67.48%	1046	9.0836	-79.6632	14-Sep-2014	this study
<i>Epiperipatus bernali</i>	141128	SRR136144 42	Panama	64.72%	904	8.4332	-82.4519	10-Sep-2014	this study
<i>Peripatus juanensis</i>	133571	SRR136144 36	Puerto Rico	6.65%	579	18.1720	-67.0445	12-Feb-2016	this study
<i>Peripatus juanensis</i>	133572	SRR136144 35	Puerto Rico	9.61%	658	18.3232	-65.8155	19-Feb-2016	this study
<i>Epiperipatus broadwayi</i>	143935	SRR136144 26	Trinidad & Tobago (Tobago)	67.69%	975	11.2869	-60.6101	26-May-2017	this study

<i>Epiperipatus trinidadensis</i>	143926	SRR13614433	Trinidad & Tobago (Trinidad)	69.02%	1048	10.7051	-61.2898	16-May-2017	this study
<i>Macroperipatus torquatus</i>	143928	SRR13614429	Trinidad & Tobago (Trinidad)	62.78%	1013	10.7459	-61.2554	16-May-2017	this study
Family Peripatopsidae									
<i>Euperipatoides kanangrensis</i>	131395	SRR13614431	Australia (NSW)	16.87%	178	-33.9833	150.1333	—	this study
Peripatopsidae sp.	141470	SRR13614440	Australia (QLD)	61.25%	898	-27.3917	152.9236	22-Oct-2016	this study
Peripatopsidae sp.	141416	SRR13614441	Australia (QLD)	67.28%	1008	-24.4130	151.0387	25-May-2015	this study
<i>Kumbadjena occidentalis</i>	141468	SRR13614430	Australia (WA)	42.43%	551	-33.9078	115.0136	10-Jan-2016	this study
<i>Occiperipatoides gilesii</i>	141469	SRR13614423	Australia (WA)	48.16%	846	-31.8667	116.0667	27-Jul-2016	this study
<i>Metaperipatus inae</i>	138078	SRR13614424	Chile	75.87%	1037	-38.0163	-73.1790	13-Nov-2014	this study
<i>Ooperipatellus viridimaculatus</i>	29203	SRR8627697	New Zealand	33.23%	708	-44.4876	168.7874	20-Jan-2014	(Laumer et al. 2019)
<i>Peripatoides aurorbis</i>	29204	SRR8627695	New Zealand	47.24%	867	-37.8419	174.7734	10-Jan-2014	(Laumer et al. 2019)
<i>Opisthopatus kwazululandi</i>	131434	SRR8318947	South Africa	82.62%	1025	-24.9370	31.3752	14-Nov-2011	(Mapalo et al. 2020)
<i>Opisthopatus highveldi</i>	131328	SRR13614447	South Africa	21.17%	257	-28.7442	31.1376	4-Apr-2001	this study
<i>Peripatopsis bolandi</i>	49527	SRR13614438	South Africa	79.75%	1005	-34.0047	18.9944	26-Dec-2014	this study
<i>Peripatopsis overbergiensis</i>	131372	SRR1145776	South Africa	51.94%	963	-33.9819	20.8231	17-Apr-2009	(Sharma et al. 2014)

Table S2. Outgroup taxa used in phylogenetic analysis of M1 and M2.

Phylum	Species	SRA number	Source
Priapulida	<i>Priapulus caudatus</i>	---	(Kocot et al. 2017)
Nematomorpha	<i>Nectonema munidae</i>	SRR8618616	(Laumer et al. 2019)
Tardigrada	<i>Echiniscus testudo</i>	SAMN10601501–SAMN10601521	(Mapalo et al. 2020)
Tardigrada	<i>Hypsibius dujardini</i>	---	(Yoshida et al. 2017)
Arthropoda	<i>Scutigera coleoptrata</i>	SRR1158078	(Fernández et al. 2014)
Arthropoda	<i>Limulus polyphemus</i>	SRR1145732	(Sharma et al. 2014)
Arthropoda	<i>Anoplodactylus insignis</i>	SRR5237777	(Fernández et al. 2016)

Table S3. Percentage of loci sampled in Neopatida-specific matrices M5–M8 across taxa. New transcriptomes in bold. See main text for details.

Species	MCZ Catalog #	Country	M5 %	M6 %	M7 %	M8 %
<i>Oroperipatus eisenii</i>	74293	Mexico	0.73	0.93	0.73	0.75
<i>Oroperipatus</i> sp.	133614	Ecuador (Galapagos)	0.27	0.66	0.28	0.26
Peripatidae sp.	141458	Colombia	0.86	0.94	0.87	0.87
<i>Epiperipatus trinidadensis</i>	143926	Trinidad & Tobago (Trinidad)	0.89	0.91	0.89	0.91
<i>Epiperipatus broadwayi</i>	143935	Trinidad & Tobago (Tobago)	0.85	0.87	0.85	0.85
<i>Macroperipatus torquatus</i>	143928	Trinidad & Tobago (Trinidad)	0.87	0.91	0.87	0.88
<i>Epiperipatus</i> sp.	136557	Brazil (Amazonas)	0.73	0.9	0.73	0.73
<i>Epiperipatus</i> sp.	141131	Brazil (Rio de Janeiro)	0.75	0.93	0.76	0.76
<i>Epiperipatus</i> sp.	141132	Brazil (Mato Grosso)	0.56	0.73	0.56	0.58
<i>Peripatus</i> sp.	46445	Guyana	0.28	0.74	0.28	0.28
<i>Peripatus juanensis</i>	133572	Puerto Rico	0.32	0.79	0.32	0.3
<i>Peripatus juanensis</i>	133571	Puerto Rico	0.2	0.58	0.2	0.19
<i>Epiperipatus</i> sp.	141126	Panama	0.8	0.86	0.8	0.8
<i>Epiperipatus vagans</i>	141130	Panama	0.76	0.84	0.77	0.79
<i>Epiperipatus bernali</i>	141128	Panama	0.78	0.82	0.79	0.78
<i>Epiperipatus solorzanoii</i>	130840	Costa Rica	0.77	0.92	0.77	0.77
<i>Macroperipatus valerioi</i>	130841	Costa Rica	0.78	0.91	0.78	0.79
<i>Macroperipatus valerioi</i>	130842	Costa Rica	0.42	0.88	0.42	0.4
<i>Epiperipatus</i> sp.	49455	Panama	0.39	0.8	0.39	0.37

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